

Morpho-phylogenetic evidence reveals novel species and new records of *Nigrograna* (Nigrogranaceae) associated with medicinal plants in Southwestern China

Hong-Zhi Du^{1,2,3,4*}, Yu-Hang Lu^{1,3,4*}, Ratchadawan Cheewangkoon^{3,4}, Jian-Kui Liu^{1,3}

¹ School of Life Science and Technology, Center for Informational Biology, University of Electronic Science and Technology of China, Chengdu 611731, Sichuan Province, China

² School of Pharmacy, Guizhou University of Traditional Chinese Medicine, Guiyang 550025, Guizhou Province, China

³ Department of Entomology and Plant Pathology, Faculty of Agriculture, Chiang Mai University, Chiang Mai 50200, Thailand

⁴ Innovative Agriculture Research Centre, Faculty of Agriculture, Chiang Mai University, Chiang Mai 50200, Thailand

Corresponding authors: Jian-Kui Liu (liujiankui@uestc.edu.cn); Ratchadawan Cheewangkoon (ratchadawan.c@cmu.ac.th)

Abstract

During a survey of saprobic fungal niches in Southwestern China, eighteen ascomycetous collections of *Nigrograna* (Nigrogranaceae, Pleosporales, Dothideomycetes) were found on dead branches of medicinal plants. These taxa were characterized and identified based on morphological and culture characteristics, and phylogenetic analyses of a combined the internal transcribed spacer region of rDNA (ITS), nuclear large subunit rDNA (28S, LSU), RNA polymerase second-largest subunit (*rpb2*), nuclear small subunit rDNA (18S, SSU), and translation elongation factor 1-alpha (*tef1-α*) sequence dataset also confirmed their placement. As a result, four novel species, namely *Nigrograna camelliae*, *N. guttulata*, *N. longiorostiolata* and *N. neriicola* were described. Additionally, four new host records of *N. acericola*, *N. magnoliae*, *N. oleae* and *N. thymi* were introduced. Furthermore, this study addresses the taxonomic status of *N. trachycarpi*, proposing its synonymy under *N. oleae*. Detailed illustrations, descriptions and informative notes for each newly identified taxon and novel host record are provided in this study.

Key words: 4 new taxa, Dothideomycetes, multi-locus, phylogeny, sexual morph, taxonomy



Academic editor: Chitrabhanu Bhunjun

Received: 20 July 2024

Accepted: 23 September 2024

Published: 23 October 2024

Citation: Du H-Z, Lu Y-H, Cheewangkoon R, Liu J-K (2024) Morpho-phylogenetic evidence reveals novel species and new records of *Nigrograna* (Nigrogranaceae) associated with medicinal plants in Southwestern China. MycoKeys 110: 1–33. <https://doi.org/10.3897/mycokeys.110.132628>

Copyright: © Hong-Zhi Du et al. This is an open access article distributed under terms of the Creative Commons Attribution License (Attribution 4.0 International – CC BY 4.0).

Introduction

The utilization of medicinal plants is integral to disease prevention and treatment in human life (Nalawade et al. 2003; Cole et al. 2007; Schmidt 2017; Rahman et al. 2019). These plants harbor diverse biological compounds that hold potential for drug development due to their rich reservoir of bioactive ingredients (Samy and Gopalakrishnakone 2007; Ali et al. 2021; Atanasov et al. 2021). Southwestern China, recognized as one of the primary regions for traditional Chinese herbal medicine, boasts remarkable diversity in medicinal plant species. This diversity is largely driven by the region's unique karst landforms, which

* These authors contributed equally to this work.

promote species differentiation and abundance (Huang et al. 2012; Taylor et al. 2014; Lu et al. 2018; Guo et al. 2020; Shan et al. 2022). Recent studies in Southwest China have revealed novel micro-fungi species associated with medicinal plants, including pathogens (Abtahi and Nourani 2017), saprobes (Du et al. 2022a; Sun et al. 2023; Wu et al. 2024) and endophytes (Helaly et al. 2018; Keshri et al. 2021; Du et al. 2022b), highlighting the potential of these plants as reservoirs for discovering fungal diversity.

Nigrogranaceae (Pleosporales, Dothideomycetes) was established by Jaklitsch and Voglmayr (2016) based on morpho-molecular evidence, with *Nigrograna* as the type genus. The divergence of Nigrogranaceae is established at approximately 79 (44–124) Mya in crown age and 131 (86–180) Mya in stem age (Liu et al. 2017). Initially, the genus *Nigrograna* was proposed by de Gruyter et al. (2012) to accommodate *Pyrenochaeta mackinnonii* (a pathogenic species in humans isolated from a mycetoma patient), which was later synonymized as *Nigrograna mackinnonii*. However, phylogenetic studies revealed that *N. mackinnonii* was closely related to *Biatrispora marina*, the type species of the monotypic genus *Biatrispora* (Ahmed et al. 2014), leading to the reclassification of *N. mackinnonii* as *Biatrispora mackinnonii* (Ahmed et al. 2014). Jaklitsch and Voglmayr (2016) subsequently proposed the family Nigrogranaceae, describing three new taxa that differed significantly from *Biatrispora* in morphology and ecology. Hongsanan et al. (2020) further revised the taxonomic status of *Biatrispora* and *Nigrograna*, suggesting that both genera should be retained. To date, there are 37 species epithets of *Nigrograna* listed in Index Fungorum (<http://www.indexfungorum.org/Names/Names.asp>; accessed 7 September 2024).

Most members of *Nigrograna* have cryptic morphological characters, leading Jaklitsch and Voglmayr (2016) to classify them as cryptic species. The sexual morphs of *Nigrograna* are characterized by having globose to subglobose ascomata with ostiole, multi-layered peridium, clavate and fissitunicate asci, fusoid to narrowly ellipsoid, straight or curved, septate, and smooth or verruculose ascospores (Jaklitsch and Voglmayr 2016; Zhang et al. 2020; Lu et al. 2022). In contrast, the asexual morphs are defined by globose to subglobose or pyriform pycnidia, filiform and branched conidiophores, hyaline, phialidic and discrete conidiogenous cells, sub-hyaline, aseptate and ellipsoidal conidia (de Gruyter et al. 2012; Jaklitsch and Voglmayr 2016; Lu et al. 2022). The life modes of *Nigrograna* species are diverse, ranging from endophytic and saprobic to pathogenic (in human) (Kolařík 2018; Zhao et al. 2018; Lu et al. 2022; Li et al. 2023). These species have been reported from various hosts in terrestrial, marine, and freshwater habitats (Hyde et al. 2017; Tibpromma et al. 2017; Dayarathne et al. 2020; Lu et al. 2022; Bundhun et al. 2023; Hu et al. 2023; Li et al. 2023; Senanayake et al. 2023; Shu et al. 2023), underscoring the broad ecological diversity of this genus. In recent years, an increasing number of new species and records of *Nigrograna* have been reported from various hosts in China. Most of these species have been identified as saprotrophic fungi from terrestrial habitats (Tibpromma et al. 2017; Boonmee et al. 2021; de Silva et al. 2022; Lu et al. 2022; Hu et al. 2023; Li et al. 2023; Liu et al. 2024; Ren et al. 2024; Xu et al. 2024). However, reports of *Nigrograna* occurring on medicinal plants are

limited. Given the ecological and economic importance of these plants, it is essential to explore the taxonomy and phylogeny of *Nigrograna* species associated with medicinal flora. Such investigations will deepen our understanding of fungal diversity in these specialized niches and may reveal new insights into the potential applications of these fungi.

This study focuses on elucidating the diversity of Nigrogranaceae in Southwestern China, identifying eight species associated with medicinal plants. We aim to describe these novel findings and contribute to the understanding of fungal diversity in this region. Through a combination of morphological comparisons and multi-locus phylogenetic analyses, we introduce four new species and four new host records, supported by both morphological and phylogenetic evidence.

Materials and methods

Collection and examination of specimens

Specimens in this study were collected from medicinal plants of nine families (Apocynaceae, Berberidaceae, Buxaceae, Celastraceae, Eucommiaceae, Fabaceae, Primulaceae, Rutaceae and Theaceae) in Southwest China during 2021 and 2023, viz., (1) Guizhou Province (26°30'43"N–26°32'18"N, 106°39'32"E–106°41'48"E, elevation 1,127–1,155 m); (2) Sichuan Province (29°29'1"N–31°8'4"N, 103°2'23"E–104°14'19"E, elevation 504–1,200 m); (3) Yunnan Province (21°55'53"N–25°14'27"N, 101°23'19"E–102°44'28"E, elevation 505–1,922 m). The sampling information (date, host, place, GPS, etc.) was recorded. Samples were packaged in envelopes and brought to the laboratory following the method described by Senanayake et al. (2020). Morphological observations were made using a Motic SMZ (Stereoscopic Zoom Microscope) 168 Series dissecting microscope (Motic, Xiamen, China) for fungal structures on a natural substrate. Fruiting bodies were collected using a syringe needle and transferred to a drop of tap water on a clean slide. The features were examined and photographed using a Nikon ECLIPSE Ni-U compound microscope fitted with a Nikon DS-Ri2 digital camera. Measurements were made with the Tarosoft Image Frame Work v. 0.9.7 software following the procedures outlined by Liu et al. (2010), and images used for photo plates were processed with Adobe Photoshop CC 2018 software (Adobe Systems, San Jose, CA, USA). Single spore isolations were made on potato dextrose agar (PDA, Oxoid) or water agar (WA, Oxoid) and later transferred onto new PDA plates following the methods described in Senanayake et al. (2020). Incubation and cultural growth were observed at 25 °C in dark and pure cultures were obtained.

Herbarium specimens were deposited in the Herbarium of Cryptogams, Kunming Institute of Botany Academia Sinica (HKAS), Kunming, China, and the herbarium of University of Electronic Science and Technology (HUEST), Chengdu, China. The pure cultures obtained in this study were deposited in the China General Microbiological Culture Collection Center (CGMCC) in Beijing, China and the University of Electronic Science and Technology Culture Collection (UESTCC), Chengdu, China. Names of the new taxa were registered in MycoBank (<http://www.mycobank.org/>).

DNA extraction, PCR amplification and sequencing

Isolates were grown in PDA medium at 25 °C in dark for three weeks to one month. Fungal mycelia were scraped off and transferred to 1.5 mL microcentrifuge tubes using a sterilized lancet for genomic DNA extraction. Fungal DNA was extracted from mycelia (about 50–100 mg) using the Trelief™ Plant Genomic DNA Kit (TsingKe Co., Beijing, China). Five different gene regions were amplified by Polymerase Chain Reaction (PCR). The internal transcribed spacer region of rDNA (ITS), nuclear large subunit rDNA (28S, LSU), nuclear small subunit rDNA (18S, SSU), RNA polymerase second-largest subunit (*rpb2*) and translation elongation factor 1- α (*tef1- α*) were selected for the study. The primers used were LR0R/LR5 for LSU (Vilgalys and Hester 1990), NS1/NS4 for SSU (White et al. 1990), ITS5/ITS4 for ITS (White et al. 1990), fRPB2-5F and fRPB2-7cR for *rpb2* (Liu et al. 1999) and TEF1-983F/TEF1-2218R for *tef1- α* (Rehner and Buckley 2005). Amplifications were performed in a 25 μ L reaction volume containing 9.5 μ L of ddH₂O, 12.5 μ L of 2 \times Taq PCR Master Mix with blue dye (Sangon Biotech, Shanghai, China), 1 μ L of DNA template and 1 μ L of each primer. The amplification condition for ITS, LSU, SSU, and *tef1- α* consisted of initial denaturation at 94 °C for 3 min, followed by 40 cycles of 45 s at 94 °C, 50 s at 55 °C and 1 min at 72 °C, and a final extension period of 10 min at 72 °C. The amplification condition for the *rpb2* gene consisted of initial denaturation at 95 °C for 5 min; followed by 37 cycles of 15 s at 95 °C, 50 s at 56 °C and 2 min at 72 °C, and a final extension period of 10 min at 72 °C. The PCR product purification and sequencing were performed at Beijing Tsingke Biotechnology (Chengdu) Co., Ltd., Chengdu, China.

Phylogenetic analyses

In this study, the taxa included in the phylogenetic analyses were selected and obtained from previous studies and GenBank (Table 1), with a total of 67 taxa. *Occultibambusa pustula* (MFLUCC 11-0502) and *O. bambusae* (MFLUCC 13-0855) (Occultibambusaceae, Pleosporales) were selected as outgroup taxa. Single-locus alignments were made in MAFFT v. 7 (<http://mafft.cbrc.jp/alignment/server/>) (Kato and Standley 2013) and checked visually using AliView (Larsson 2014). The alignments were trimmed using trimAl v 1.2 (Capella-Gutiérrez et al. 2009). Five single-locus alignments were combined using SequenceMatrix 1.7.8 (Vaidya et al. 2011). Maximum likelihood (ML) and Bayesian inference (BI) analyses were employed to assess phylogenetic relationships as detailed in Dissanayake et al. (2020).

ML analyses were performed with RAxML-HPC v.8 on XSEDE (8.2.12) (Stamatakis 2006; Stamatakis et al. 2008) through the CIPRES Science Gateway V. 3.3 (<https://www.phylo.org/portal2/login!input.action>) (Miller et al. 2010). The tree search included 1,000 non-parametric bootstrap replicates; the best scoring tree was selected among suboptimal trees from each run by comparing likelihood scores under the GTRGAMMA substitution model. The resulting replicates were plotted onto the best scoring tree obtained previously. ML bootstrap values equal to or greater than 75% were marked near each node.

BI was performed in MrBayes 3.2.6 (Ronquist et al. 2012). The program MrModeltest 2 v. 2.3 (Nylander 2008) was used to determine the best nucleotide

substitution model for each data partition. The evolutionary model of SYM+I+G substitution model was selected for ITS, HKY+G substitution model was selected for SSU, and GTR+I+G substitution model was selected for LSU, *rpb2* and *tef1-α*. Posterior probabilities (PP) (Rannala and Yang 1996) were determined by Markov chain Monte Carlo sampling (MCMC). Six simultaneous Markov chains were run for 10 million generations, and trees were sampled every 1,000 th generation. The first 25% of saved trees, representing the burn-in phase of the analysis, were discarded. The remaining trees were used for calculating posterior probabilities in the majority rule consensus tree (Larget and Simon 1999). PP values equal to or greater than 0.95 were marked near each node.

Phylogenetic trees were printed with Fig Tree v. 1.4.4 (<http://tree.bio.ed.ac.uk/software/figtree/>) and the layout was created in Adobe Illustrator CS6 software (Adobe Systems, USA). The new sequences generated in this study were deposited in GenBank (Table 1).

Table 1. Taxa used in the phylogenetic analyses and the corresponding GenBank accession numbers.

Taxa names	Strain/Specimen number	GenBank accession numbers					References
		ITS	LSU	<i>rpb2</i>	SSU	<i>tef1-α</i>	
<i>Nigrograna acericola</i>	CGMCC 3.24957 [†]	OR253153	OR253312	N/A	N/A	OR263572	Li et al. (2023)
<i>Nigrograna acericola</i>	UESTCC 23.0208	PP812425	PP812460	PP838917	PP812443	PP838935	In this study
<i>Nigrograna acericola</i>	UESTCC 23.0191	PP812426	PP812461	PP838918	PP812444	PP838936	In this study
<i>Nigrograna antibiotica</i>	CCF 4378 [†]	JX570932	KF925327	N/A	KF925328	JX570934	Kolařík (2018)
<i>Nigrograna antibiotica</i>	CCF 4498	LT221894	LT221895	N/A	N/A	N/A	Kolařík (2018)
<i>Nigrograna aquatica</i>	MFLUCC 17-2318 [†]	MT627705	MN913705	N/A	N/A	N/A	Dong et al. (2020)
<i>Nigrograna asexualis</i>	ZHKUCC 22-0214 [†]	OP450965	OP450971	OP432241	OP450979	OP432245	Lu et al. (2022)
<i>Nigrograna camelliae</i>	CGMCC 3.25625[†]	PP812431	PP812466	PP838923	PP812449	PP838939	In this study
<i>Nigrograna camelliae</i>	UESTCC 23.0197	PP812432	PP812468	PP838924	PP812450	PP838940	In this study
<i>Nigrograna cangshanensis</i>	MFLUCC 15-0253 [†]	KY511063	KY511064	N/A	KY511065	N/A	Tibpromma et al. (2017)
<i>Nigrograna carollii</i>	CCF 4484 [†]	LN626657	LN626682	LN626662	LN626674	LN626668	Kolařík (2018)
<i>Nigrograna chromolaenae</i>	MFLUCC 17-1437 [†]	MT214379	MT214473	N/A	N/A	MT235801	Mapook et al. (2020)
<i>Nigrograna coffeae</i>	ZHKUCC 22-0210 [†]	OP450967	OP450973	OP432243	OP450981	OP432247	Lu et al. (2022)
<i>Nigrograna coffeae</i>	ZHKUCC 22-0211	OP450968	OP450974	OP432244	OP450982	OP432248	Lu et al. (2022)
<i>Nigrograna fuscidula</i>	CBS 141556 [†]	KX650550	N/A	N/A	N/A	KX650525	Jaklitsch and Voglmayr (2016)
<i>Nigrograna fuscidula</i>	CBS 141476	KX650547	N/A	KX650576	KX650509	KX650522	Jaklitsch and Voglmayr (2016)
<i>Nigrograna guizhouensis</i>	CGMCC 3.25501 [†]	OR680498	OR680565	OR842915	OR680867	OR858897	Zhang et al. (2024)
<i>Nigrograna guizhouensis</i>	ZY22.020	OR680499	OR680566	OR842916	OR680868	OR858898	Zhang et al. (2024)
<i>Nigrograna guttulata</i>	CGMCC 3.25689[†]	PP812433	PP812469	PP838925	PP812451	PP838941	In this study
<i>Nigrograna guttulata</i>	UESTCC 23.0295	PP812434	PP812470	PP838926	PP812452	PP838942	In this study
<i>Nigrograna heveae</i>	ZHKUCC 22-0284 [†]	OP584490	OP584488	OP750374	OP584492	OP750372	Hyde et al. (2023)
<i>Nigrograna hydei</i>	GZCC 19-0050 [†]	MN387225	MN387227	N/A	N/A	MN389249	Zhang et al. (2020)
<i>Nigrograna impatientis</i>	GZCC 19-0042 [†]	MN387226	MN387228	N/A	N/A	MN389250	Zhang et al. (2020)
<i>Nigrograna italica</i>	MFLU 23-0139 [†]	OR538590	OR538591	OR531365	N/A	OR531366	Bundhun et al. (2023)
<i>Nigrograna jinghongensis</i>	KUMUCC 21-0035 [†]	MZ493303	MZ493317	MZ508421	MZ493289	MZ508412	Boonmee et al. (2021)
<i>Nigrograna jinghongensis</i>	KUMUCC 21-0036	MZ493304	MZ493318	MZ508422	MZ493290	MZ508413	Boonmee et al. (2021)
<i>Nigrograna kunmingensis</i>	ZHKUCC 22-0242 [†]	OP456214	OP456379	N/A	OP456382	OP471608	Liu et al. (2024)
<i>Nigrograna kunmingensis</i>	ZHKUCC 22-0243	OP484334	OP456380	N/A	OP456383	OP471609	Liu et al. (2024)
<i>Nigrograna lincangensis</i>	ZHKUCC 23-0798 [†]	OR853099	OR922323	OR966280	OR941079	OR966282	Xu et al. (2024)
<i>Nigrograna lincangensis</i>	ZHKUCC 23-0799	OR853100	OR922324	OR966281	OR941080	OR966283	Xu et al. (2024)

Taxa names	Strain/Specimen number	GenBank accession numbers					References
		ITS	LSU	<i>rpb2</i>	SSU	<i>tef1-a</i>	
<i>Nigrograna locuta-pollinis</i>	CGMCC 3.18784 ^T	MF939601	MF939583	MF939610	N/A	MF939613	Zhao et al. (2018)
<i>Nigrograna longiorostiolata</i>	CGMCC 3.25626^T	PP812421	PP812458	PP838913	PP812439	PP838945	In this study
<i>Nigrograna longiorostiolata</i>	UESTCC 23.0200	PP812422	PP812457	PP838914	PP812440	PP838946	In this study
<i>Nigrograna mackinnonii</i>	CBS 674.75 ^T	KF015654	KF015612	KF015703	GQ387552	KF407986	de Gruyter et al. (2012)
<i>Nigrograna magnoliae</i>	MFLUCC 20-0020 ^T	MT159628	MT159622	MT159611	MT159634	MT159605	Wanasinghe et al. (2020)
<i>Nigrograna magnoliae</i>	MFLUCC 20-0021	MT159629	MT159623	MT159612	MT159635	MT159606	Wanasinghe et al. (2020)
<i>Nigrograna magnoliae</i>	UESTCC 23.0203	PP812419	PP812454	PP838929	PP812437	PP838943	In this study
<i>Nigrograna magnoliae</i>	CGMCC 3.25627	PP812420	PP812453	PP838927	PP812435	PP838931	In this study
<i>Nigrograna magnoliae</i>	UESTCC 23.0190	PP812417	PP812456	PP838930	PP812438	PP838944	In this study
<i>Nigrograna magnoliae</i>	UESTCC 23.0206	PP812418	PP812455	PP838928	PP812436	PP838932	In this study
<i>Nigrograna mycophila</i>	CBS 141478 ^T	KX650553	N/A	N/A	N/A	KX650526	Jaklitsch and Voglmayr (2016)
<i>Nigrograna mycophila</i>	CBS 141483	KX650555	N/A	KX650577	KX650510	KX650528	Jaklitsch and Voglmayr (2016)
<i>Nigrograna neriicola</i>	CGMCC 3.25624^T	PP812430	PP812467	PP838921	PP812447	PP838937	In this study
<i>Nigrograna neriicola</i>	UESTCC 23.0195	PP812429	PP812465	PP838922	PP812448	PP838938	In this study
<i>Nigrograna norvegica</i>	CBS 141485 ^T	KX650556	N/A	KX650578	KX650511	N/A	Jaklitsch and Voglmayr (2016)
<i>Nigrograna obliqua</i>	CBS 141477 ^T	KX650560	N/A	KX650580	N/A	KX650531	Jaklitsch and Voglmayr (2016)
<i>Nigrograna obliqua</i>	CBS 141475	KX650558	N/A	KX650579	KX650512	KX650530	Jaklitsch and Voglmayr (2016)
<i>Nigrograna oleae</i>	CGMCC 3.24423 ^T	OR253080	OR253232	N/A	N/A	OR262140	Li et al. (2023)
<i>Nigrograna oleae</i> (<i>N. trachycarpi</i>)	GMB0499	OR120437	N/A	N/A	N/A	OR150024	Hu et al. (2023); In this study
<i>Nigrograna oleae</i> (<i>N. trachycarpi</i>)	GMB0505	OR120440	N/A	N/A	N/A	OR150025	Hu et al. (2023); In this study
<i>Nigrograna oleae</i>	UESTCC 23.0209	PP812424	PP812463	PP838915	PP812441	PP838933	In this study
<i>Nigrograna oleae</i>	UESTCC 23.0193	PP812423	PP812459	PP838916	PP812442	PP838934	In this study
<i>Nigrograna peruviansis</i>	CCF 4485 ^T	LN626658	LN626683	LN626665	LN626677	LN626671	Kolařík (2018)
<i>Nigrograna puerensis</i>	ZHKUCC 22-0212 ^T	OP450969	OP450975	N/A	OP450983	OP432249	Lu et al. (2022)
<i>Nigrograna rhizophorae</i>	MFLUCC 18-0397 ^T	MN047085	N/A	MN431489	N/A	MN077064	Dayarathne et al. (2020)
<i>Nigrograna rubescens</i>	CHEM 2344 ^T	OQ400924	OQ400934	OQ413082	N/A	OQ413077	Mack et al. (2024)
<i>Nigrograna samueliana</i>	NFCCI 4383 ^T	MK358817	MK358812	MK330939	MK358810	MK330937	Dayarathne et al. (2020)
<i>Nigrograna schinifolii</i>	GMB0498 ^T	OR120434	N/A	N/A	N/A	OR150022	Hu et al. (2023)
<i>Nigrograna schinifolii</i>	GMB0504	OR120441	N/A	N/A	N/A	OR150023	Hu et al. (2023)
<i>Nigrograna sichuanensis</i>	CGMCC 3.24424 ^T	OR253096	OR253248	N/A	N/A	OR251058	Li et al. (2023)
<i>Nigrograna thailandica</i>	MFLUCC 17-2663	MK762709	MK762716	N/A	MK762704	N/A	Senanayake et al. (2023)
<i>Nigrograna thymi</i>	MFLUCC 14-1096 ^T	KY775576	KY775573	N/A	KY775574	KY775578	Hyde et al. (2017)
<i>Nigrograna thymi</i>	UESTCC 23.0210	PP812428	PP812464	PP838919	PP812445	N/A	In this study
<i>Nigrograna thymi</i>	UESTCC 23.0194	PP812427	PP812462	PP838920	PP812446	N/A	In this study
<i>Nigrograna verniciae</i>	CGMCC 3.24425	OR253116	OR253275	N/A	N/A	OR251168	Li et al. (2023)
<i>Nigrograna wuhanensis</i>	ZHKUCC 22-0329 ^T	OP941389	OP941390	N/A	OQ061465	OP947079	Shu et al. (2023)
<i>Nigrograna yasuniana</i>	YU 101026 ^T	HQ108005	LN626684	LN626664	LN626676	LN626670	Kolařík (2018)
<i>Occultibambusa bambusae</i>	MFLUCC 13-0855 ^T	KU940123	KU863112	KU940170	N/A	KU940193	Dai et al. (2017)
<i>Occultibambusa pustula</i>	MFLUCC 11-0502 ^T	KU940126	KU863115	N/A	N/A	N/A	Dai et al. (2017)

*** Remarks:** The superscript T denotes ex-type isolates. “N/A” denotes sequence is unavailable. The newly generated sequences, new species and synonymized isolates are indicated in black bold font. **Abbreviations:** **CBS:** CBS–KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; **CCF:** Culture Collection of Fungi, Charles University, Prague, Czech Republic; **CGMCC:** China General Microbiological Culture Collection Center, Institute of Microbiology, Chinese Academy of Sciences, Beijing, China; **GMB:** Herbaria of Guizhou Medical University, Guiyang, China; **GZCC:** Guizhou Culture Collection, Guizhou, China; **KUMUCC:** Kunming Medical University Culture Collection, Kunming, China; **MFLUCC:** Mae Fah Luang University Culture Collection, Chiang Rai, Thailand; **NFCCI:** National Fungal Culture Collection of India, India; **UESTCC:** University of Electronic Science and Technology Culture Collection, Chengdu, China; **YU:** Yale University Herbarium, Connecticut, America; **ZHKUCC:** Zhongkai University of Agriculture and Engineering Culture Collection, Guangzhou, China; **Personal collections:** **ZY** and **CHEM:** These numbers are assigned by the author and have no annotations.

Phylogenetic results

In this study, five loci, ITS, LSU, *rpb2*, SSU, and *tef1-α*, were used to determine the phylogenetic placement of the new collections. The concatenated matrix was comprised of 69 taxa with a total of 4,236 bp characters (ITS: 1–473 bp; LSU: 474–1,306 bp; *rpb2*: 1,307–2,331 bp; SSU: 2,332–3,335 bp; *tef1-α*: 3,336–4,236 bp) including gaps. Single-locus analyses were carried out to compare the topologies and clade stabilities, respectively. The results showed that ML and BI were similar in topology without significant conflicts. The best RAxML tree with a final likelihood value of -20,464.246121 is presented in Fig. 1. RAxML analysis yielded 1,200 distinct alignment patterns and 26.42% of undetermined characters or gaps. Estimated base frequencies were as follows: A = 0.247595, C = 0.247214, G = 0.264771, T = 0.240420, with substitution rates AC = 1.651782, AG = 5.572876, AT = 1.491922, CG = 1.179397, CT = 11.546850, GT = 1.000000; gamma distribution shape parameter alpha = 0.139617. Tree-Length = 1.453662. The final average standard deviation of split frequencies at the end of total MCMC generations for BI analysis was 0.009978 (the critical value for the topological convergence diagnostic is below 0.01).

Representatives of all the species of *Nigrograna* were including in our phylogenetic analysis (Fig. 1). Four strains (CGMCC 3.25627, UESTCC 23.0203, UESTCC 23.0190 and UESTCC 23.0206) were nested with *N. magnoliae* (ex-type strain MFLUCC 20-0020 and MFLUCC 20-0021), and strains, UESTCC 23.0208 and UESTCC 23.0191, UESTCC 23.0210 and UESTCC 23.0194, clustered with *N. acericola* (ex-type strain, CGMCC 3.24957) and *N. thymi* (ex-type strain, MFLUCC 14-1096), respectively. *Nigrograna trachycarpi* (GMB0499 and GMB0505) was synonymized under *N. oleae*, these two strains of *N. trachycarpi* and our two isolates (UESTCC 23.0209 and UESTCC 23.0193) grouped with ex-type strain of *N. oleae* (CGMCC 3.24423) with maximum support (100% MLBS/1.00 BIPP).

Nigrograna camelliae (CGMCC 3.25625 and UESTCC 23.0197) and *N. guttulata* (CGMCC 3.25689 and UESTCC 23.0295) were sister to *N. coffeae* (ex-type strain ZHKUCC 22-0210 and ZHKUCC 22-0211) and *N. peruviansis* (ex-type strain CCF 4485), respectively. They formed two distinct clades with 100% MLBS/1.00 BIPP and 63% MLBS/0.98 BIPP, respectively. *Nigrograna neriicola* (CGMCC 3.25624 and UESTCC 23.0195) was sister to *N. schinifolii* (ex-type strain GMB0498 and GMB0504) and formed a strongly supported monophyletic lineage (96% MLBS/1.00 BIPP). *Nigrograna longiorostiolata* (CGMCC 3.25626 and UESTCC 23.0200) formed a distinct lineage with high bootstrap support (100% MLBS/1.00 BIPP).

Taxonomy

Nigrograna magnoliae Wanas, PLoS One, 15(7): 10 (2020)

MycoBank No: 557331

Fig. 2

Description. *Saprobic* on dead branches of *Buxus sinica* (Buxaceae). **Sexual morph:** *Ascomata* 204–326 μm wide, 140–220 μm high (\bar{x} = 248 × 187 μm, n = 20), solitary or gregarious, scattered, immersed to semi-immersed, with only ostiolar necks visible on the host surface, trigonoid, uniloculate, perithecioid, globose to subglobose, brown to dark brown, with an ostiole. **Ostiole** central or eccentric,

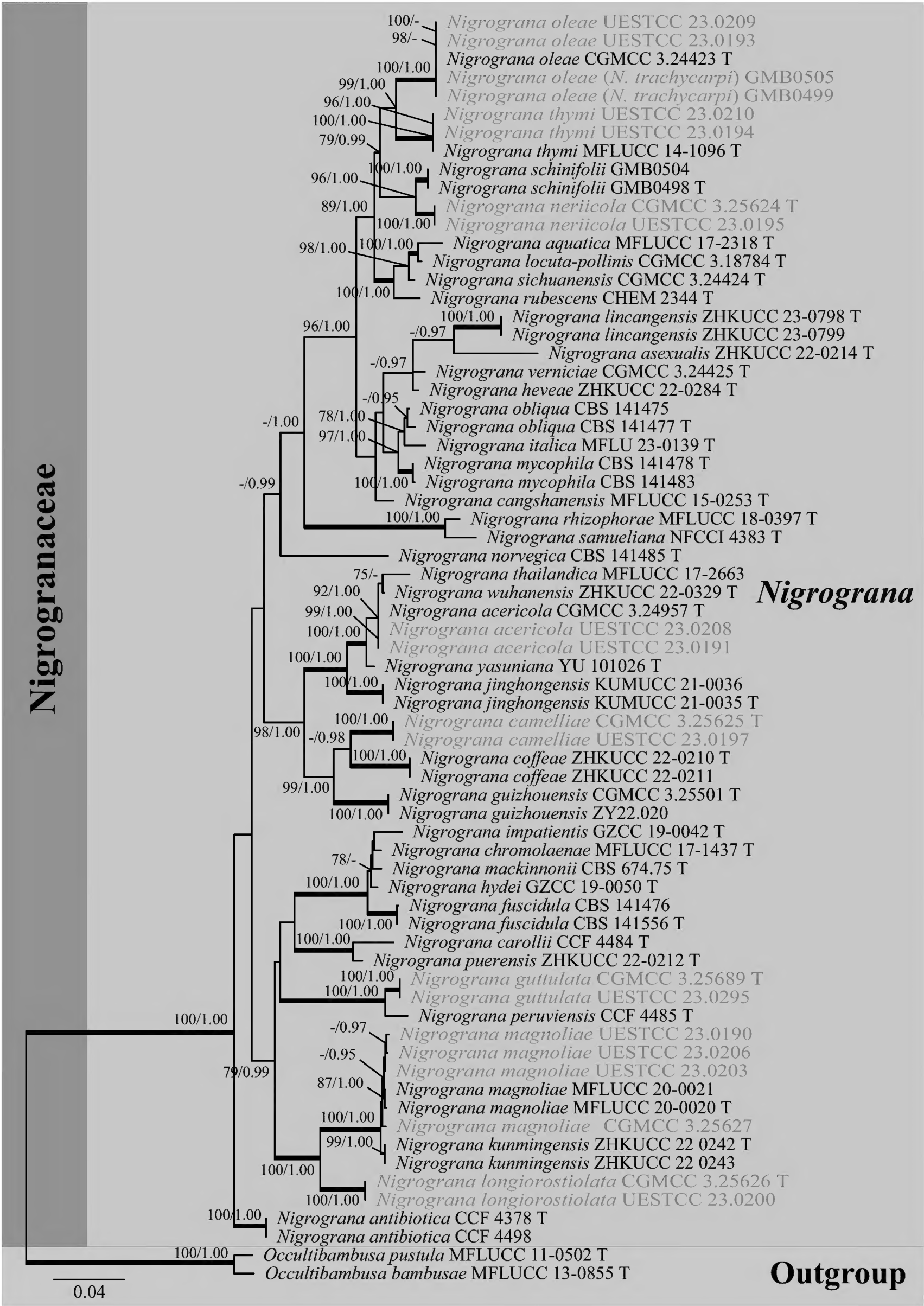


Figure 1. Phylogenetic tree constructed from maximum likelihood (RAxML) analyses of a combined ITS, LSU, *rpb2*, SSU, and *tef1-α* sequence data for selected genera within the family Nigrogranaceae (Pleosporales, Dothideomycetes). Branches support for Maximum likelihood (MLBS) equal to or greater than 75% and Bayesian inference posterior probabilities (BIPP) equal to or greater than 0.95 are marked above or below nodes as MLBS/BIPP. The abbreviation T indicates the ex-type strain. Species names and culture collections in red are newly collected taxa and synonymized isolates. The tree was rooted with *Occultibambusa pustula* (MFLUCC 11-0502) and *O. bambusae* (MFLUCC 13-0855).

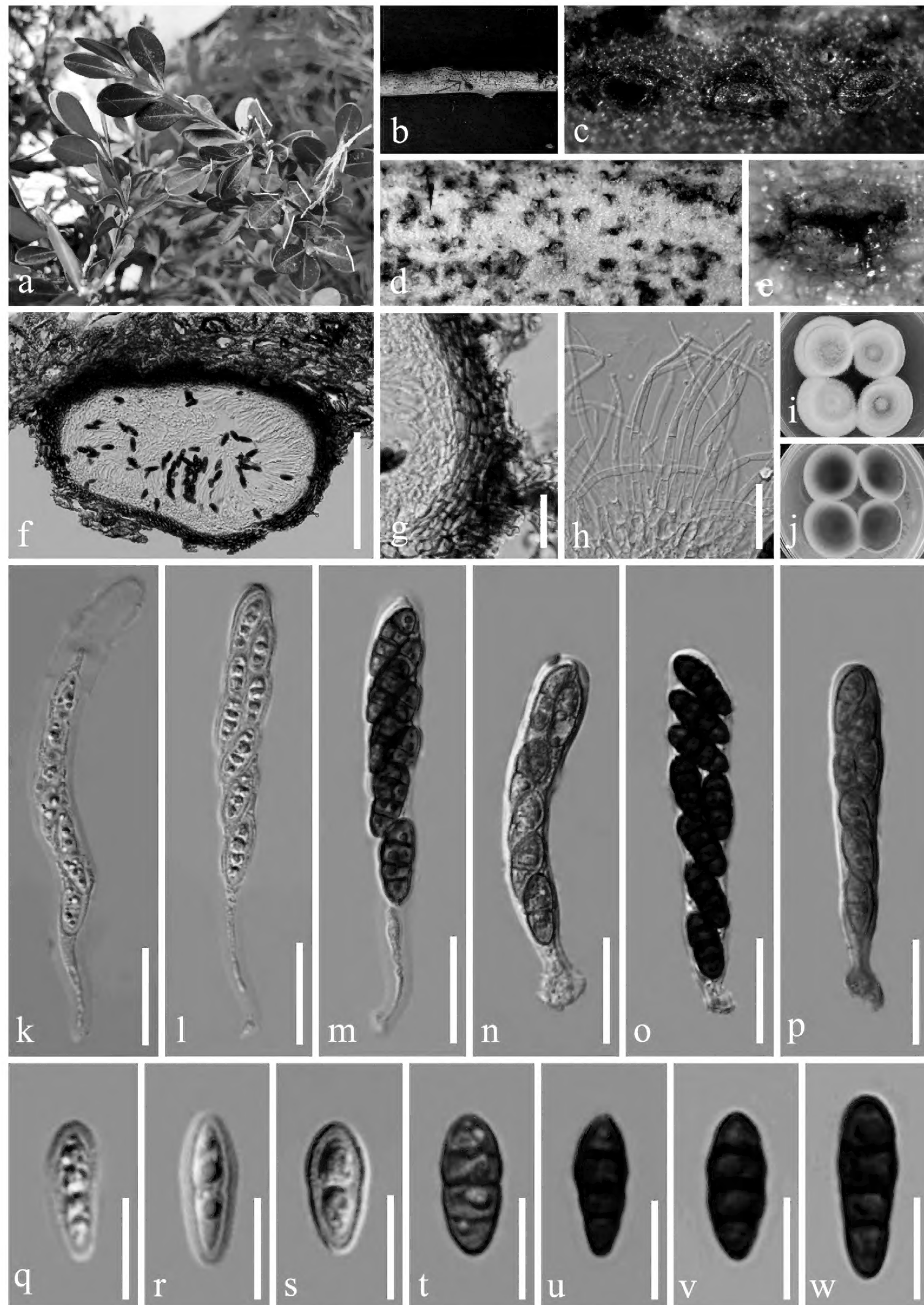


Figure 2. *Nigrograna magnoliae* (HUEST 23.0203, new host record) **a** host *Buxus sinica* **b** branch of *Buxus sinica* **c–e** appearance of ascomata on host surface **f** vertical section through ascoma **g** peridium **h** hamathecium **i, j** colonies on PDA, above (**i**) and below (**j**) **k–o** asci **p** asci in Congo red **q–w** ascospores. Scale bars: 100 µm (**f**); 20 µm (**g, h, k–p**); 10 µm (**q–w**).

brittle. **Peridium** 15–23 µm (\bar{x} = 18 µm, n = 20) composed of angular cells, consisting 4–5 layers, brown to dark brown thick-walled cells of outer layer, hyaline to subhyaline thin-walled cells of inner layer. **Hamathecium** 1–3 µm (\bar{x} = 2 µm, n = 20) wide, composed of numerous, filamentous, hyaline, aseptate or separate, rarely branched, smooth-walled pseudoparaphyses. **Asci** 57–103 × 8–11 µm (\bar{x} = 72.5 × 10 µm, n = 30), 8-spored, bitunicate, fissitunicate, clavate to long cylindric-clavate, short cylindrical pedicellate with a swollen base, apically rounded, with a minute

ocular chamber. **Ascospores** 13–19 × 4.5–6 µm (\bar{x} = 14.5 × 5 µm, n = 50), 1–2-se-riate, partially overlapping, fusoid to ellipsoid, tapering towards the blunt ends, or blunt at both ends, guttulate, smooth-walled, olivaceous to yellowish-brown when young, 1-septate; deeply constricted at septa, becoming 3-septate, brown to dark brown when mature, without appendages. **Asexual morph**: Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 32–33 mm in diameter after three weeks at 25 °C in dark, white in the whole colony from above, and slightly raised in the center, circular, flat, edge entire, margin well-defined; in reverse, grayish black in the center, off-white at the margin, the color gradually lightens from center to edge, no pigmentation on PDA.

Material examined. CHINA • Yunnan Province, Kunming City, Panlong District, Kunming Botanical Garden. 25°8'27"N, 102°44'24"E, elevation 1,922 m, on dead branches of medicinal plant *Buxus sinica* (Rehder & E. H. Wilson) M. Cheng (Buxa-ceae), 11 November 2022, H.Z. Du, S735 (HUEST 23.0203), living culture UESTCC 23.0203; • *ibid.*, Sichuan Province, Chengdu City, High-tech West District, Yaobo Park, 30°43'57"N, 103°56'21"E, elevation 504 m, on dead branches of medicinal plant *Eucommia ulmoides* Oliv. (Eucommiaceae), 11 August 2021, H.Z. Du, S347 (HUEST 23.0206), living culture UESTCC 23.0206; • *ibid.*, Guizhou Province, Guiyang City, Nanming District, Guiyang Medicinal Botanical Garden, 26°32'18"N, 106°41'48"E, elevation 1,127 m, on dead branches of medicinal plant *Mahonia bealei* (Fort.) Carr. (Berberidaceae), 12 October 2021, H.Z. Du, S370 (HUEST 23.0207), living culture CGMCC 3.25627 = UESTCC 23.0207; • *ibid.*, Guizhou Prov-ince, Guiyang City, Huaxi District, 26°30'43"N, 106°39'32"E, elevation 1,155 m, on dead branches of medicinal plant *Camellia sinensis* (L.) O. Ktze. (Theaceae), 2 February 2023, Y.X. Yu, GY33 (HUEST 23.0190), living culture UESTCC 23.0190.

Notes. *Nigrograna magnoliae* was introduced by Wanasinghe et al. (2020) with both asexual and sexual morphs reported in China. The host distribution of this species is presented in Table 2. Our collections are identical to *N. magnoli-ae* based on morphology and phylogeny. Therefore, we reported it as new host records from medicinal plants of *Buxus sinica*, *Camellia sinensis*, *Eucommia ulmoides* and *Mahonia bealei* in China.

Table 2. The host distribution of *Nigrograna magnoliae*.

Host distribution	Collecting sites	References
<i>Magnolia denudate</i> (Magnoliaceae)	China (Yunnan Province)	Wanasinghe et al. (2020)
Submerged wood from aquatic habitats	Thailand (Chiang Rai Province)	Zhang et al. (2020)
Decaying twigs of unidentified host	China (Guizhou Province)	Zhang et al. (2020)
<i>Acer truncatum</i> (Aceraceae)	China (Sichuan Province)	Li et al. (2023)
<i>Juglans regia</i> (Juglandaceae)	China (Sichuan Province)	Li et al. (2023)
<i>Olea europaea</i> (Oleaceae)	China (Sichuan Province)	Li et al. (2023)
<i>Michelia alba</i> (Magnoliaceae)	China (Guizhou Province)	Chethana et al. (2023)
<i>Rosa</i> sp. (Rosaceae)	China (Sichuan Province)	Chethana et al. (2023)
Fruiting bodies of <i>Shearia</i> sp. (Dothioraceae)	China (Guizhou Province)	Chethana et al. (2023)
<i>Magnolia grandiflora</i> (Magnoliaceae)	Thailand (Chiang Mai Province)	https://www.ncbi.nlm.nih.gov/nuccore/MN081891.1
<i>Castanopsis indica</i> (Fagaceae)	China (Yunnan Province)	Ren et al. (2024)
<i>Buxus sinica</i> (Buxaceae)	China (Yunnan Province)	In this study
<i>Eucommia ulmoides</i> (Eucommiaceae)	China (Sichuan Province)	In this study
<i>Mahonia bealei</i> (Berberidaceae)	China (Guizhou Province)	In this study
<i>Camellia sinensis</i> (Theaceae)	China (Guizhou Province)	In this study

***Nigrograna longiorostiolata* H.Z. Du & Jian K. Liu, sp. nov.**

MycoBank No: 854177

Fig. 3

Etymology. The epithet '*longiorostiolata*' refers to the longer-ostiolate of ascomata.

Holotype. HKAS 131311

Description. **Saprobic** on dead branches of *Citrus medica* (Rutaceae). **Sexual morph: Ascomata** 222–293 µm wide, 144–486 µm high (\bar{x} = 264 × 303 µm, n = 20), solitary, scattered, immersed, visible as black dots on the host surface, uniloculate, globose to subglobose, sometimes obpyriform with a long ostiole. **Ostioles** 175–302 µm long, 83–128 µm wide (\bar{x} = 263 × 102 µm, n = 20) central or eccentric, longer, with a crest-like apex, filled with hyaline or slightly brown periphyses. **Peridium** 17–32 µm (\bar{x} = 23.5 µm, n = 20) composed of *textura prismatica* cells, consisting 3–4 layers, brown to dark brown of outer layer, hyaline to subhyaline of inner layer. **Hamathecium** 1–2 µm (\bar{x} = 1.5 µm, n = 20) wide, composed of numerous, filiform, hyaline, aseptate or separate, rarely branched, guttulate, smooth-walled pseudoparaphyses. **Asci** 40–70 × 6–9 µm (\bar{x} = 53 × 8 µm, n = 30), 5–8-spored, bitunicate, fissitunicate, clavate, short cylindrical pedicellate with a swollen base, apically rounded, with a minute ocular chamber. **Ascospores** 10–13 × 4–6 µm (\bar{x} = 12 × 5 µm, n = 50), 1–2-seriate, partially overlapping, fusoid to ellipsoid, tapering towards the blunt ends, or blunt at both ends, guttulate, olivaceous to yellowish-brown when young, aseptate or 1-septate; deeply constricted at septa, becoming 3-septate, brown to dark brown when mature, without appendages. **Asexual morph:** Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 17–18 mm in diameter after three weeks at 25 °C in dark, white in the whole colony from above, circular, edge entire, margin well-defined; in reverse, off-white to grayish brown, no pigmentation on PDA.

Material examined. CHINA • Yunnan Province, Xishuangbanna Dai Autonomous Prefecture, Mengla County, Xishuangbanna Tropical Botanical Garden Chinese Academy of Sciences. 21°56'1"N, 101°25'33"E, elevation 505 m, on dead branches of medicinal plant *Citrus medica* L. (Rutaceae), 10 November 2022, H.Z. Du, S655 (HKAS 131311, holotype; HUEST 23.0200, isotype); ex-holotype living culture CGMCC 3.25626; ex-isotype living culture UESTCC 23.0200.

Notes. *Nigrograna longiorostiolata* shares similar morphology with *N. magnoliae* (holotype, MFLU 20–0092) and *N. kunmingensis* (holotype, ZHKU 22–0141) in having immersed, globose to subglobose ascomata, bitunicate and clavate asci, fusoid to ellipsoid, 3-septate mature ascospores. However, the ascomata size of *N. longiorostiolata* (222–293 × 144–486 µm) is larger than *N. magnoliae* (200–300 × 100–150 µm) (Wanasinghe et al. 2020) and smaller than *N. kunmingensis* (300–500 × 390–450 µm) (Liu et al. 2024). The phylogenetic result (Fig. 1) showed that *N. longiorostiolata* (CGMCC 3.25626 and UESTCC 23.0200) can be recognized as a distinct phylogenetic species with high bootstrap support (100% MLBS/1.00 BIPP). Additionally, *N. longiorostiolata* (ex-type strain, CGMCC 3.25626) can be distinguished from *N. magnoliae* (ex-type strain, MFLUCC 20-0020) by 26/471 bp (5.5%, 2 gaps) in ITS, 14/831 bp (1.7%, without gaps) in LSU, 30/855 bp (3.5%, 3 gaps) in *tef1-α* and 96/1042

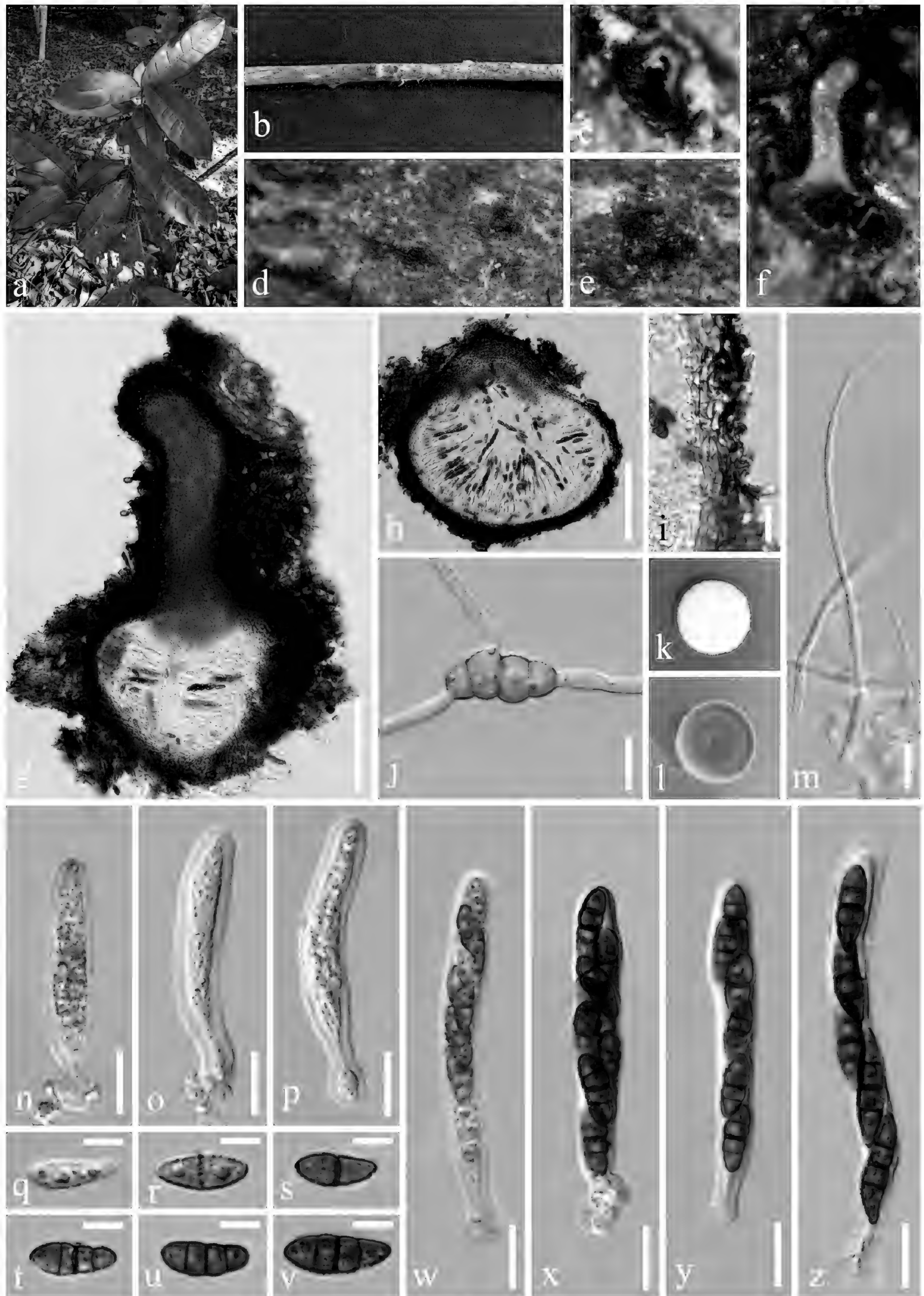


Figure 3. *Nigrograna longiorostiolata* (HKAS 131311, holotype) **a** host *Citrus medica* **b** branch of *Citrus medica* **c–f** appearance of ascomata on host surface **g, h** vertical section through ascoma **i** peridium **j** germinated ascospore **k, l** colony on PDA, above (**k**) and below (**l**) **m** hamathecium **n–p, w–z** asci **q–v** ascospores. Scale bars: 100 μm (**g, h**); 10 μm (**i, j, m–p, w–z**); 5 μm (**q–v**).

bp (9.2%, without gaps) in *rpb2* differences, and differs from *N. kunmingensis* (ex-type strain, ZHKUCC 22-0242) with 70/823 bp (8.5%, 21 gaps) of ITS, 14/844 bp (1.7%, without gaps) of LSU and 30/855 bp (3.5%, 3 gaps) of *tef1-a* differences. Therefore, *N. longiorostiolata* associated with *Citrus medica* is a phylogenetically distinct specie and introduced as a new species.

***Nigrograna acericola* W.L. Li & Jian K. Liu, Mycosphere, 14(1): 1496–1500 (2023)**

MycoBank No: 849155

Fig. 4

Description. *Saprobic* on dead branches of *Gymnosporia acuminata* (Celastraceae). **Sexual morph:** **Ascomata** 524–647 × 341–475 µm (\bar{x} = 586 × 424 µm, n = 20), solitary, scattered, immersed, ostiolar necks visible on the host surface or erumpent, subglobose to ellipsoid, coriaceous, brown to dark brown, with an ostiole. **Ostioles** 86–138 µm long, 64–119 µm wide (\bar{x} = 113 × 96 µm, n = 20), mostly central, some eccentric, with a crest-like apex, central, filled with hyaline periphyses. **Peridium** 15–58 µm (\bar{x} = 40 µm, n = 20) µm wide, composed of 4–5 layers of flattened, brown to dark brown, thin-walled cells of *textura angularis*, the inner layer is dense, the outer layer sparse. **Hamathecium** 1.5–3 µm (\bar{x} = 2 µm, n = 20) wide, composed of numerous, filamentous, hyaline, unbranched pseudoparaphyses. **Asci** 70–87 × 12–14 µm (\bar{x} = 77 × 13 µm, n = 30), 8-spored, bitunicate, fissitunicate, cylindrical to clavate, short pedicellate, apically rounded, with a minute ocular chamber. **Ascospores** 16–19 × 5–7 µm (\bar{x} = 17 × 6 µm, n = 50), 1–2-seriate, biseriate or partially overlapping, fusoid to ellipsoid, with obtuse ends, tapering towards the ends, guttulate, smooth-walled, 1-septate, subhyaline to yellowish-brown when young; becoming 3-septate, slightly constricted at the middle septum, brown to dark brown when mature, without appendages. **Asexual morph:** Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 38–40 mm in diameter after three weeks at 25 °C in dark, white in the whole colony from above, circular, edge entire, margin well-defined; in reverse, light brown in the center, olive gray at the margin, no pigmentation on PDA.

Material examined. CHINA • Yunnan Province, Xishuangbanna Dai Autonomous Prefecture, Mengla County, Xishuangbanna Tropical Botanical Garden Chinese Academy of Sciences. 21°55'54"N, 101°15'16"E, elevation 511 m, on dead branches of medicinal plant *Gymnosporia acuminata* Hook. f. (Celastraceae), 10 November 2022, H.Z. Du, D03 (HUEST 23.0208), living culture UESTCC 23.0208; *ibid.*, Sichuan Province, Zigong City, Rong County, 29°29'1"N, 104°14'19"E, elevation 850 m, on dead branches of medicinal plant *Camellia sinensis* (L.) O. Ktze. (Theaceae), 3 November 2022, Y. H. Lu & Y. Xiao, CS11 (HUEST 23.0191), living culture UESTCC 23.0191.

Notes. *Nigrograna acericola* was introduced by Li et al. (2023) from *Acer truncatum* (Aceraceae) in China. Our collections are identical to *N. acericola* based on morphology and phylogeny. We reported it as new host records from *Camellia sinensis* and *Gymnosporia acuminata* in China.

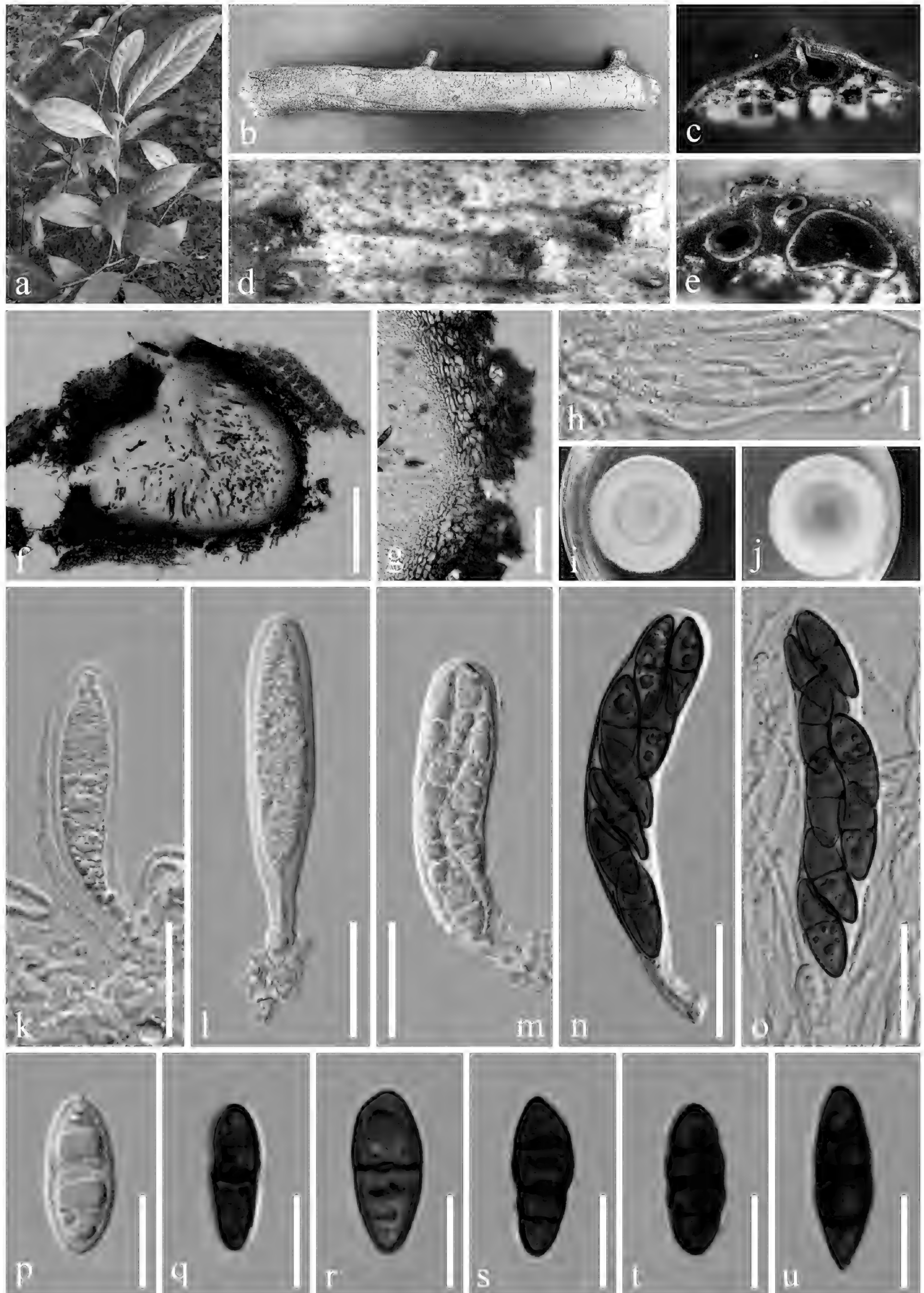


Figure 4. *Nigrograna acericola* (HUEST 23.0208, new host record) **a** host *Gymnosporia acuminata* **b** branch of *Gymnosporia acuminata* **c–e** appearance of ascomata on host surface **f** vertical section through ascoma **g** peridium **h** hamathecium **i, j** colony on PDA, above (**i**) and below (**j**) **k–o** asci. **p–u** ascospores. Scale bars: 200 μm (**f**); 50 μm (**g**); 10 μm (**h, p–u**); 20 μm (**k–o**).

***Nigrograna camelliae* Y.H. Lu, H.Z. Du & Jian K. Liu, sp. nov.**

MycoBank No: 854178

Fig. 5

Etymology. The epithet '*camelliae*' refers to the host genus *Camelliae* from which the fungus was originally isolated.

Holotype. HKAS 131310

Description. **Saprobic** on dead branches of *Camellia sinensis* (Theaceae). **Sexual morph: Ascomata** 137–270 µm wide, 208–324 µm high (\bar{x} = 212 × 265 µm, n = 20), solitary, scattered, immersed, black spots on the host substrate, globose to subglobose, sometimes obpyriform, ostiolate, hairs of ascomata 2–3 µm wide, slightly brown, septate. **Ostioles** 65–138 µm long, 32–60 µm wide (\bar{x} = 100 × 45 µm, n = 20) mostly central, some eccentric, with a crest-like apex. **Peridium** 19–30 µm (\bar{x} = 23 µm, n = 20) wide, composed of 2–3 layers, comprising reddish brown to dark brown pigmented cells. **Hamathecium** 2–3 µm (\bar{x} = 2.5 µm, n = 20) wide, composed of numerous, filiform, hyaline, aseptate or separate, filamentous, smooth-walled pseudoparaphyses. **Asci** 70–108 × 9–11 µm (\bar{x} = 80 × 10 µm, n = 30), 8-spored, bitunicate, fissitunicate, clavate to cylindric-clavate, short stalked, some with a swollen base, apically rounded, with a small ocular chamber. **Ascospores** 13–16 × 4–6 µm (\bar{x} = 15 × 5 µm, n = 50), overlapping uni- to bi-seriately arranged, fusoid to ellipsoid, tapering towards the blunt ends, or blunt at both ends, straight or slightly curved, 1-septate, constricted, with obviously guttulate, hyaline to slightly brown when immature, pale brown to brown when mature, without appendages. **Asexual morph:** Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 35–36 mm in diameter after three weeks at 25 °C in dark, white in the whole colony and slightly bright yellow in the center from above, circular, edge entire, margin well-defined; in reverse, yellowish brown in the center, slightly brown at the margin and presented an outer ring, no pigmentation on PDA.

Material examined. CHINA • Sichuan Province, Yaan City, Mingshan County, Mengding Mountain. 30°4'32"N, 103°2'23"E, elevation 1,200 m, on dead branches of medicinal plant *Camellia sinensis* (L.) O. Ktze. (Theaceae), 16 July 2023, Y. H. Lu & X. D. Liang, MD03A (HKAS 131310, holotype; HUEST 23.0197, isotype); ex-holotype living culture CGMCC 3.25625; ex-isotype living culture UESTCC 23.0197.

Notes. *Nigrograna camelliae* is phylogenetically close to *N. coffeae* and represents as a distinct lineage (Fig. 1). Additionally, the nucleotide base pair comparison between *N. camelliae* (ex-type strain, CGMCC 3.25625) and *N. coffeae* (ex-type strain, ZHKUCC 22-0210) revealed 15/514 bp (2.9%, 1 gap) of ITS, 11/698 bp (1.6%, without gaps) of LSU, 74/739 bp (10.0% without gaps) of *rpb2* and 28/914 bp (3.1%, without gaps) of *tef1-α* differences. Furthermore, *N. camelliae* morphologically resembles *N. coffeae* in having immersed ascomata, clavate and short pedicellate asci, pale brown to brown and septate ascospores with obviously guttulate (Lu et al. 2022). However, *N. camelliae* differs from *N. coffeae* in having ascomata with hairs and ostioles, solitary or scattered in the substrate. Additionally, they can be distinguished in having larger ascomata (208–324 × 137–270 µm vs. 140–200 × 90–140 µm) and asci (70–108 × 9–11 µm vs. 50–70 × 7–11 µm). Therefore, *N. camelliae* is introduced as a new species with the justification of phylogenetic and morphological evidence.

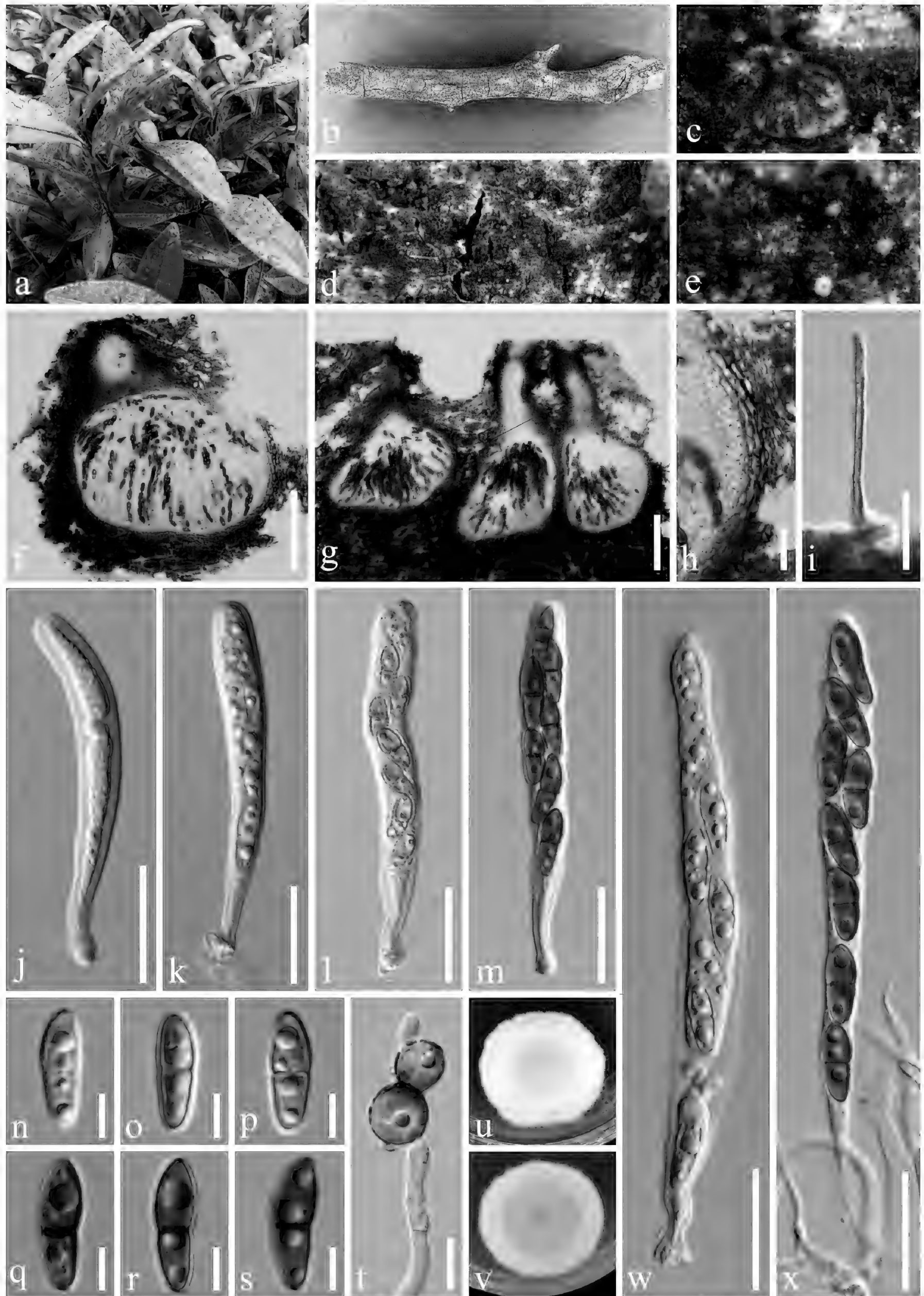


Figure 5. *Nigrograna camelliae* (HKAS 131310, holotype) **a** host *Camellia sinensis* **b** branch of *Camellia sinensis* **c–e** appearance of ascomata on host surface **f, g** vertical section through ascoma **h** peridium **i** hairs on ascomata **j–m, w, x** asci **n–s** ascospores **t** germinated ascospore **u, v** colony on PDA, above (**u**) and below (**v**). Scale bars: 100 µm (**f, g**); 20 µm (**h–m, w, x**); 5 µm (**n–s**); 10 µm (**t**).

***Nigrograna oleae* W.L. Li & Jian K. Liu, Mycosphere, 14(1): 1503–1505 (2023)**

MycoBank No: 849157

Fig. 6

= *Nigrograna trachycarpi*, MycoKeys 100: 141 (2023).

Description. *Saprobic* on dead branches of *Ardisia crenata* (Primulaceae). **Sexual morph:** **Ascomata** 190–334 µm wide, 303–406 µm high (\bar{x} = 233 × 370 µm, n = 20), solitary or gregarious, scattered, immersed, often lying parallelly or obliquely to the bark or host surface, with a cylindrical ostiolar neck, coriaceous, obpyriform, brown to dark brown. **Ostioles** central or eccentric, filled with hyaline periphyses. **Peridium** 16.5–25 µm (\bar{x} = 21 µm, n = 20) wide, consisting 4–6 layers of brown-walled cells of **textura angularis**. **Hamathecium** 1–2 µm (\bar{x} = 1.5 µm, n = 20) wide, aseptate or separate, composed of numerous, filiform, smooth-walled pseudoparaphyses. **Asci** 62–127 × 9–12 µm (\bar{x} = 82 × 10 µm, n = 30), 8-spored, bitunicate, fissitunicate, clavate to long cylindric-clavate, with a short pedicel, apically rounded, with a smaller ocular chamber. **Ascospores** 14–17 × 4–6 µm (\bar{x} = 15 × 5 µm, n = 50), 1–2-seriate, fusoid to ellipsoid, apical cell mostly obtuse, straight or slightly curved, guttulate, smooth-walled, 3-septate, constricted at the septa, pale brown to yellow-brown when young, brown to chocolate-brown at maturation, without appendages. **Asexual morph:** Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 22–23 mm in diameter after three weeks at 25 °C in dark. Colonies from above, circular, margin entire, dense, surface smooth, velvety appearance, white in the center, presented a pale greenish furrowed ring, white to cream at the margin; in reverse, brown in the central point, brown-gray in the middle, white to pale brownish at the edge, no pigmentation on PDA.

Material examined. CHINA • Yunnan Province, Xishuangbanna Dai Autonomous Prefecture, Mengla County, Xishuangbanna Tropical Botanical Garden Chinese Academy of Sciences. 21°55'49"N, 101°15'19"E, elevation 516 m, on dead branches of medicinal plant *Ardisia crenata* Sims (Primulaceae), 10 November 2022, H.Z. Du, D01 (HUEST 23.0209), living culture UESTCC 23.0209; • *ibid.*, Sichuan Province, Chengdu City, Pujiang County, 30°11'42"N, 103°22'21"E, elevation 630 m, on dead branches of *Camellia sinensis* (L.) O. Ktze. (Theaceae), 5 October 2022, Y.H. Lu & Y. Xiao, A11 (HUEST 23.0193), living culture UESTCC 23.0193.

Notes. *Nigrograna oleae* was introduced by Li et al. (2023) from *Olea europaea* and *N. trachycarpi* was described by Hu et al. (2023) from *Trachycarpus* sp. in China. In this study, multi-locus phylogeny indicated that our two isolates clustered together with *N. oleae* (ex-type strain, CGMCC 3.24423) and *N. trachycarpi* (ex-type strain, GMB0499) by strong support (100% MLBS/1.00 BIPP) (Fig. 1). In addition, the nucleotide base pair comparison of ex-type strain between *N. oleae* (CGMCC 3.24423) and *N. trachycarpi* (GMB0499) was identical by 421/421 bp (100%) of ITS, and 466/466 bp (100%) of *tef1-α*. Additionally, our newly collected specimens share similar morphology with *N. oleae* and *N. trachycarpi*. Therefore, we identify our collections as *N. oleae* and propose the synonymy of *N. trachycarpi* under *N. oleae* based on morphology and phylogeny. The new host records for *N. oleae* from medicinal plants *Ardisia crenata* and *Camellia sinensis* are reported in this study.



Figure 6. *Nigrograna oleae* (HUEST 23.0209, new host record) **a** host *Ardisia crenata* **b** branch of *Ardisia crenata* **c–f** appearance of ascomata on host surface **g, h** vertical section through ascoma **i** peridium **j–l** asci **m–q** ascospores **r, s** colony on PDA, above (**r**) and below (**s**). Scale bars: 100 µm (**g, h**); 20 µm (**i–l**); 10 µm (**m–q**).

***Nigrograna thymi* Mapook, Camporesi & K.D. Hyde, Fungal Diversity, 87: 68–70 (2017)**

MycoBank No: 552958

Fig. 7

Description. *Saprobic* on dead branches of *Huangtcia renifolia* (Fabaceae). **Sexual morph: Ascomata** 292–359 µm wide, 166–278 µm high (\bar{x} = 327 × 218 µm, n = 20), solitary or scattered, immersed or semi-immersed to slightly erumpent through host tissue, coriaceous, globose to subglobose, brown to dark brown, hairs of ascomata 2–3 µm wide, brown, septate, branched. **Ostiole** inconspicuous, without papillate. **Peridium** 15–44 µm (\bar{x} = 29.5 µm, n = 20) wide, 5–6 layers, comprising dark brown cells of *textura angularis*. **Hamathecium** comprising 1–3 µm (\bar{x} = 2 µm, n = 20) wide, cylindrical to filiform, septate, branched, smooth-walled pseudoparaphyses. **Asci** 43–86 × 7–9 µm (\bar{x} = 66 × 8 µm, n = 30), 8-spored, bitunicate, cylindrical to broadly filiform, with small ocular chamber. **Ascospores** 11–15 × 4–6 µm (\bar{x} = 13 × 4.5 µm, n = 50), 1–2-seriate, overlapping, broadly fusiform to inequilateral, widest at the middle cell, guttulate, smooth-walled, aseptate or 1-septate, hyaline when immature, becoming 3-septate, slightly constricted at the septum, pale brown to brown at maturity, without appendages. **Asexual morph:** Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 20 mm in diameter after three weeks at 25 °C in dark. Colonies from above, white in the whole colony and raised in the center, circular, edge entire, margin well-defined; in reverse, grayish-green in the center, white to pale green ring at the margin, no pigmentation on PDA.

Material examined. CHINA • Yunnan Province, Xishuangbanna Dai Autonomous Prefecture, Mengla County, Xishuangbanna Tropical Botanical Garden Chinese Academy of Sciences. 21°55'50"N, 101°15'29"E, elevation 515 m, on dead branches of medicinal plant *Huangtcia renifolia* (L.) H. Ohashi & K. Ohashi (Fabaceae), 10 November 2022, H.Z. Du, D02 (HUEST 23.0210), living culture UESTCC 23.0210; • *ibid.*, Sichuan Province, Leshan City, Emeishan County, 29°36'10"N, 103°21'54"E, elevation 1,100 m, on dead branches of *Camellia sinensis* (L.) O. Ktze. (Theaceae), 18 July 2023, Y.H. Lu & X.D. Liang, EM03 (HUEST 23.0194), living culture UESTCC 23.0194.

Notes. *Nigrograna thymi* was introduced by Hyde et al. (2017) from *Thymus oenipontanus* in Italy. Our collections are identical to *N. thymi* based on morphology and phylogeny (Fig. 1). We reported it as new host records from medicinal plants *Huangtcia renifolia* and *Camellia sinensis* in China.

***Nigrograna neriicola* Y.H. Lu, H.Z. Du & Jian K. Liu, sp. nov.**

MycoBank No: 854179

Fig. 8

Etymology. The epithet '*neriicola*' refers to the host genus *Nerium* from which the fungus was originally isolated.

Holotype. HKAS 131313.

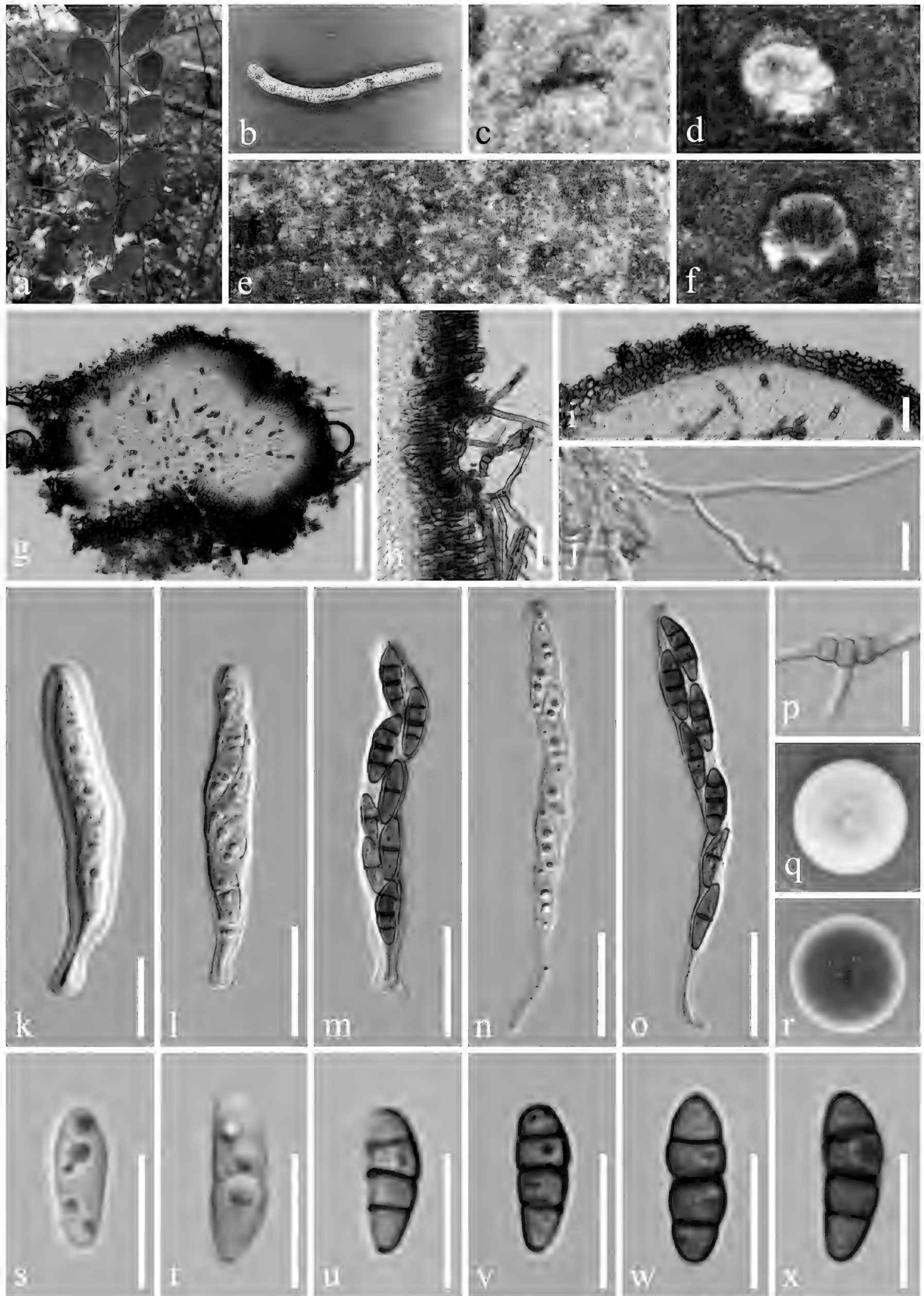


Figure 7. *Nigrograna thymi* (HUEST 23.0210, new host record) **a** host *Huangtcia renifolia* **b** branch of *Huangtcia renifolia* **c–f** appearance of ascomata on host surface **g** vertical section through ascoma **h** hairs on ascomata **i** peridium **j** hamathecium **k–o** asci **p** germinated ascospore **q, r** colony on PDA, above (**q**) and below (**r**) **s–x** ascospores. Scale bars: 100 μm (**g**); 20 μm (**h, i, l–p**); 10 μm (**j, k, s–x**).

Description. *Saprobic* on dead branches of *Nerium oleander* (Apocynaceae). **Sexual morph:** **Ascomata** 138–231 μm wide, 156–251 μm high (\bar{x} = 182 \times 202 μm , n = 20), mostly gregarious, sometimes solitary, scattered, immersed to semi-immersed, appearing as black irregular protrusions and cracks, globose to subglobose, sometimes obpyriform, dark brown to black, with an ostiole. **Ostioles** 32–54 μm long, 14–34 μm wide (\bar{x} = 45 \times 25 μm , n = 20) mostly central, some eccentric, with a crest-like apex. **Peridium** 16–61 μm (\bar{x} = 32 μm , n = 20) wide, multi-layered, reticulate structure, comprising dark brown to reddish brown pigmented cells of *textura angularis*. **Hamathecium** 1–2.5 μm wide (\bar{x} = 2 μm , n = 20), composed of numerous, filiform, hyaline, aseptate or separate, rarely branched, filamentous, smooth-walled pseudoparaphyses. **Asci** 35–80 \times 7–10 μm (\bar{x} = 56 \times 8.5 μm , n = 30), 8-spored, bitunicate, fissitunicate, clavate to cylindric-clavate, short stalked, some with club-shape pedicel, apically rounded with a small ocular chamber. **Ascospores** 12–21(–31) \times 3.5–5 μm (\bar{x} = 16 \times 4 μm , n = 50), uni- to bi-seriately arranged, partially overlapping, fusoid to ellipsoid, tapering towards the blunt ends, or blunt at both ends, straight or slightly curved, guttulate, smooth-walled, 1-septate, subhyaline to slightly brown when young; becoming 3-septate, yellowish-brown to dark brown when mature, deeply constricted at septa, without appendages. **Asexual morph:** Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 33–35 mm in diameter after one month at 25 °C in dark, slightly brown in the whole colony and raised in the central point from above, circular, edge entire, margin well-defined, aerial mycelia dense; in reverse, black-brown in the center, slightly brown ring at the margin, no pigmentation on PDA.

Material examined. CHINA • Yunnan Province, Xishuangbanna Dai Autonomous Prefecture, Mengla County, Xishuangbanna Tropical Botanical Garden Chinese Academy of Sciences. 21°55'52"N, 101°15'29"E, elevation 505 m, on dead branches of medicinal plant *Nerium oleander* L. (Apocynaceae), 10 November 2022, H.Z. Du, D04 (HKAS 131313, holotype); ex-holotype living culture CGMCC 3.25624; • *ibid.*, Sichuan Province, Chengdu City, Pujiang County. 30°11'40"N, 103°22'25"E, elevation 600 m, on dead branches of *Camellia sinensis* (L.) O. Ktze. (Theaceae), 5 October 2022, Y.H. Lu & Y. Xiao, M03 (HUEST 23.0195, paratype); ex-paratype living culture UESTCC 23.0195.

Notes. *Nigrograna neriicola* (CGMCC 3.25624 and UESTCC 23.0195) has close phylogenetic relationships with *N. schinifolii* (GMB0498 and GMB0504) but formed a distinct lineage (Fig. 1). Morphologically, the ascomata of *N. neriicola* differs from *N. schinifolii* in having black irregular protrusions and cracks, mostly gregarious, and ascospores that are slightly larger than *N. schinifolii* (12–21 \times 3.5–5 μm vs. 10–14 \times 2.8–4 μm) (Hu et al. 2023). Additionally, the nucleotide base pair comparison between *N. neriicola* (ex-type strain, CGMCC 3.25624) and *N. schinifolii* (ex-type strain, GMB0498) revealed no significant differences by 375/377 bp (99.5%, 1 gap) of ITS and 507/511 bp (99.2%, without gaps) of *tef1-a*. However, for *tef1-a* gene, the length of the two *N. schinifolii* strains (GMB0498 and GMB0504) is only 511 bp. The problem of low similarity occurred after the blastn search without a corresponding sequence in the same genus for alignment. Therefore, *N. neriicola* is introduced as a new species with the morpho-molecular data analysis.

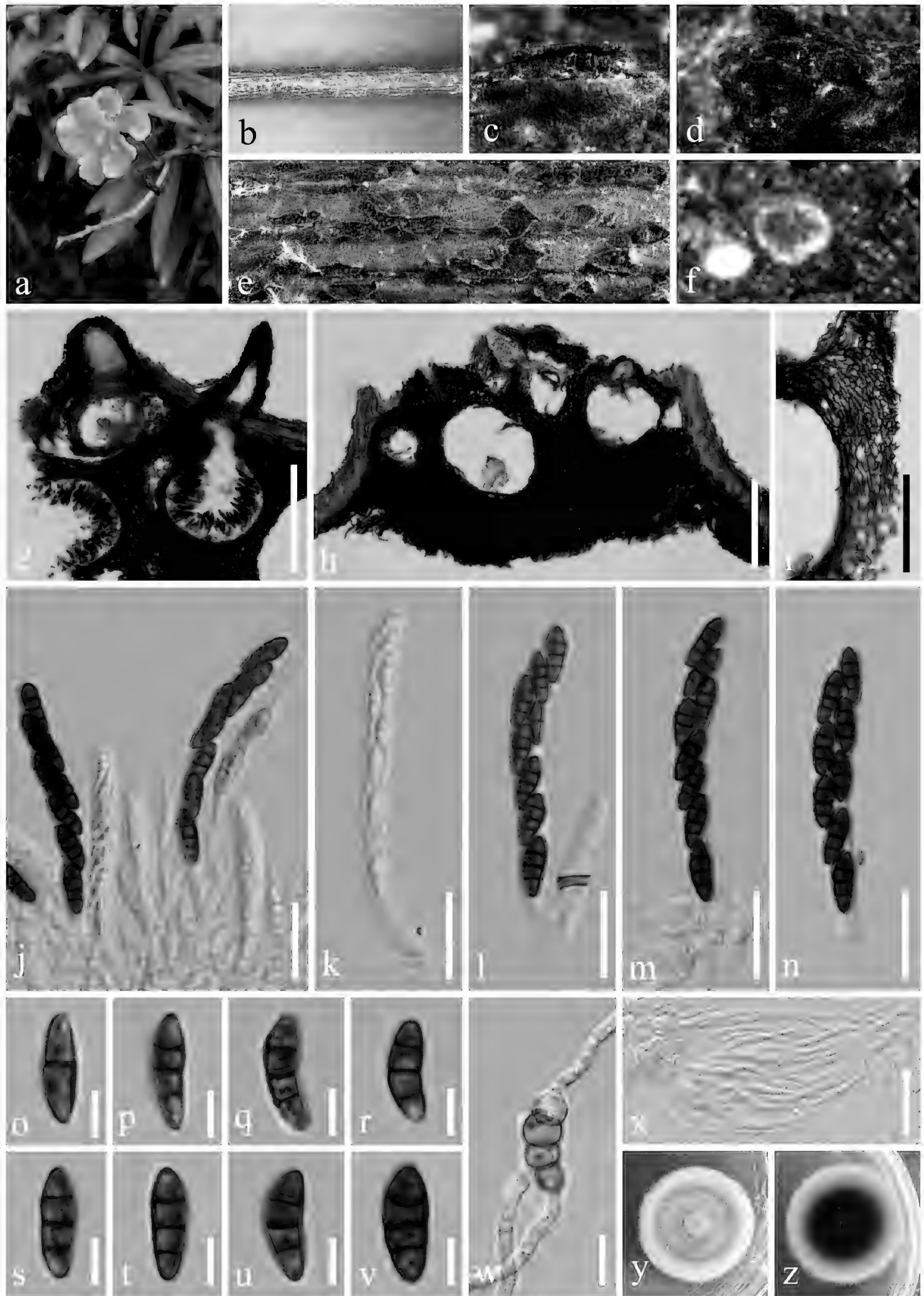


Figure 8. *Nigrograna neriicola* (HKAS 131313, holotype) **a** host *Nerium oleander* **b** branch of *Nerium oleander* **c–f** appearance of ascomata on host surface **g, h** vertical section through ascoma **i** peridium **j–n** asci **o–v** ascospores **w** germinated ascospore **x** hamathecium **y, z** colony on PDA, above (**y**) and below (**z**). Scale bars: 200 μm (**g, h**); 100 μm (**i**); 20 μm (**j–n, x**); 5 μm (**o–v**); 10 μm (**w**).

***Nigrograna guttulata* Y.H. Lu, H.Z. Du & Jian K. Liu, sp. nov.**

MycoBank No: 854180

Fig. 9

Etymology. The epithet '*guttulata*' refers to the guttulate ascospores.

Holotype. HKAS 131992.

Description. *Saprobic* on dead branches of *Camellia sinensis* (Theaceae).

Sexual morph: Ascomata 182–283 µm wide, 106–276 µm high (\bar{x} = 241 × 183 µm, n = 20), solitary, immersed, ostiolar necks visible on the host surface or erumpent, triangular, globose to subglobose, sometimes obpyriform, coriaceous, ostiolate, dark brown to black. **Ostioles** 35–61 µm long, 15–30 µm wide (\bar{x} = 47 × 22 µm, n = 20) mostly central, some eccentric, filled with hyaline periphyses. **Peridium** 15–37 µm (\bar{x} = 25 µm, n = 20) wide, multi-layered, reticulate structure, comprising dark brown to reddish brown pigmented cells of *textura angularis*. **Hamathecium** 1–2.5 µm wide (\bar{x} = 2 µm, n = 20), composed of numerous, filiform, hyaline, aseptate or separate, some branched, filamentous, smooth-walled pseudoparaphyses. **Asci** 35–70 × 7–12 µm (\bar{x} = 48 × 8.5 µm, n = 30), 8-spored, bitunicate, fissitunicate, clavate to cylindric-clavate, short stalked, some with club-shape pedicel, apically rounded, with small ocular chamber. **Ascospores** 10–13 × 3–5 µm (\bar{x} = 12 × 4 µm, n = 50), 1–2-seriate, overlapping, fusoid to ellipsoid, tapering towards the blunt ends, or blunt at both ends, straight or slightly curved, guttulate, smooth-walled, subhyaline to slightly brown when young, 1-septate; yellowish-brown to dark brown when mature, becoming 3-septate, deeply constricted at septa, without appendages.

Asexual morph: Undetermined.

Culture characteristics. Ascospores germinated on PDA within 24 h, and germ tubes produced from basal cell. Colonies growing on PDA reached 35–38 mm in diameter after one month at 25 °C in dark. Colonies from above, white in the whole colony and raised in the central point, circular, margin well-defined, aerial mycelia dense; in reverse, grayish green in the center, white ring at the margin, no pigmentation on PDA.

Material examined. CHINA • Guizhou Province, Guiyang City, Huaxi District, 26°30'40"N, 106°39'30"E, elevation 1,155 m, on dead branches of medicinal plant *Camellia sinensis* (Linnaeus) Kuntze (Theaceae), 2 February 2023, Y.X. Yu & Y.H. Lu, GY15 (HKAS 131992, holotype; HUEST 23.0295, isotype), ex-holotype living culture CGMCC 3.25689; ex-isotype living culture UESTCC 23.0295.

Notes. *Nigrograna peruviansis* was reported by Kolařík et al. (2017) as an endophytic fungus (*Biatriospora peruviansis*) and was synonymized under the genus *Nigrograna* by Kolařík (2018), but with a lack of detailed morphological structures. In this study, our isolates of *N. guttulata* (CGMCC 3.25689 and UESTCC 23.0295) have a close phylogenetic relationship with *N. peruviansis* (Kolařík et al. 2017; Kolařík 2018) based on ITS, LSU, *rpb2*, SSU, and *tef1-α* sequence data, and formed a distinct lineage with absolute bootstrap support (100% MLBS/1.00 BIPP) (Fig. 1). Additionally, *N. guttulata* (ex-type strain, CGMCC 3.25689) can be distinguished from *N. peruviansis* (ex-type strain, CCF 4485) by 8/462 bp (1.7%, 3 gaps) in ITS, 24/1020 bp (2.4%, without gaps) in LSU and 10/618 bp (1.6%, without gaps) in *rpb2* differences. Therefore, the establishment of the new species *N. guttulata* is justified by the phylogenetic evidence.

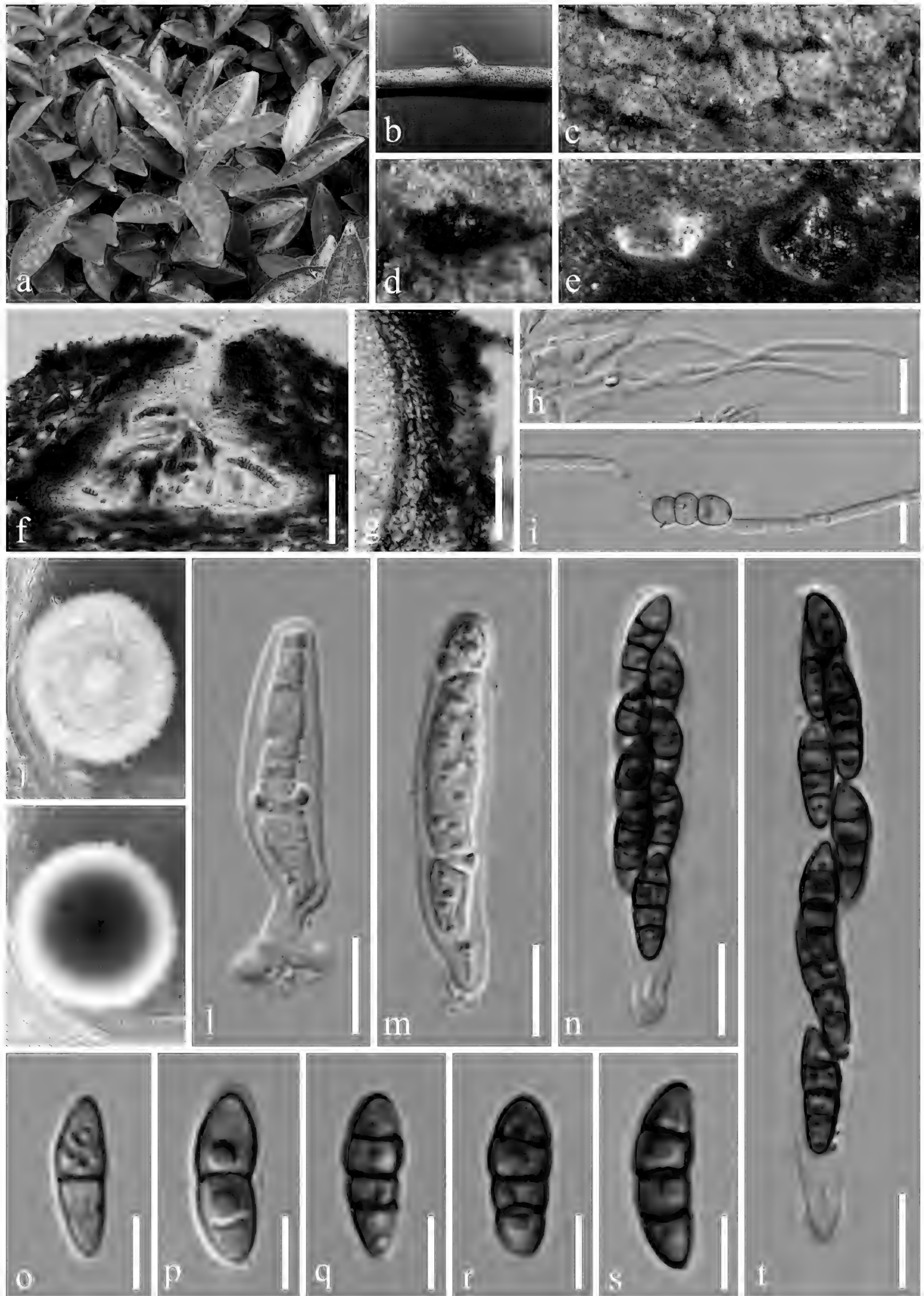


Figure 9. *Nigrograna guttulata* (HKAS 131992, holotype) **a** host *Camellia sinensis* **b** branch of *Camellia sinensis* **c–e** appearance of ascomata on host surface **f** vertical section through ascoma **g** peridium **h** hamathecium **i** germinated ascospore **j, k** colony on PDA, above (**j**) and below (**k**) **l–n, t** asci **o–s** ascospores. Scale bars: 50 μm (**f**); 40 μm (**g**); 10 μm (**h, i, l–n, t**); 5 μm (**o–s**).

Discussion

In this study, eighteen isolates of *Nigrograna* (Nigrogranaceae, Pleosporales, Dothideomycetes) were obtained from medicinal plants in Southwest China (Guizhou, Sichuan and Yunnan Provinces). Based on morphological and culture characteristics, and phylogenetic analyses of combined ITS, LSU, *rpb2*, SSU, and *tef1-α* sequence data, four novel species were identified, namely *Nigrograna camelliae*, *N. guttulata*, *N. longiorostiolata* and *N. neriicola*. Additionally, our known species, namely *N. acericola*, *N. magnoliae*, *N. oleae* and *N. thymi*, were reported from medicinal plants as new host records. These isolates were associated with terrestrial habitat and collected from medicinal plants in nine plant families, including Apocynaceae, Berberidaceae, Buxaceae, Celastraceae, Eucommiaceae, Fabaceae, Primulaceae, Rutaceae, and Theaceae.

Species within the genus *Nigrograna* exhibit considerable morphological similarity, often complicating species delimitation based solely on morphological traits (Jaklitsch and Voglmayr 2016; Zhang et al. 2020; Lu et al. 2022; Li et al. 2023). As such, molecular data play a critical role in species identification. For example, Jaklitsch and Voglmayr (2016) demonstrated that morphologically similar species, such as *N. coffeae* and *N. camelliae*, can be distinguished phylogenetically. These species sequence divergence across multiple loci, including (15/514 bp, 2.9%, 1 gap), LSU (11/698 bp, 1.6%, without gaps), *rpb2* (74/739 bp, 10.0%, without gaps) and *tef1-α* (28/914 bp, 3.1%, without gaps), highlighting the importance of molecular analysis for accurate taxonomic placement. *Nigrograna* is a worldwide distributed genus, with species reported from Asia (Dayarathne et al. 2020; Mapook et al. 2020; Zhang et al. 2020; Lu et al. 2022; Li et al. 2023), the Americas, and Europe (Jaklitsch and Voglmayr 2016; Hyde et al. 2017; Kolařík et al. 2017; Tibpromma et al. 2017; Kolařík 2018; Zhao et al. 2018; Dayarathne et al. 2020; Wanasinghe et al. 2020). While certain species, such as *N. carollii*, *N. peruviansis* and *N. yasuniana*, have been reported as endophytes on various hosts (Kolařík et al. 2017), the majority of known species are saprotrophs on the bark or corticated twigs and branches of various hardwoods (Jaklitsch and Voglmayr 2016; Mapook et al. 2020; Zhang et al. 2020; Lu et al. 2022; Hu et al. 2023; Li et al. 2023). Reports of *Nigrograna* species on flowers, fruits, leaves, or herbaceous plants are rare, indicating a preference for woody hosts. Consistent with these findings, the isolates in this study were primarily recovered from the branches of medicinal woody plants, such as *Eucommia ulmoides* (Eucommiaceae), *Gymnosporia acuminata* (Celastraceae), and *Mahonia bealei* (Berberidaceae).

It is noteworthy that *Nigrograna magnoliae* was isolated from the bark of *Eucommia ulmoides*, which is a primary medicinal component of the plant. The quality of medicinal plants is closely tied to their clinical efficacy, and the presence of fungal species such as *N. magnoliae* raises important questions about the potential impact of fungal colonization on the medicinal properties of their hosts (Balekundri and Mannur 2020; Rasool et al. 2020; Ali et al. 2021). This finding warrants further investigation to assess whether *N. magnoliae* could affect the quality or bioactive compounds of *E. ulmoides*. In addition to their ecological diversity, certain species within *Nigrograna* have been found to produce bioactive secondary metabolites. For instance, *Nigrograna rubescens* has been reported to produce naphthoquinone compounds, which are known for their broad spectrum

of biological activities (Naysmith et al. 2017; Mack et al. 2024). These metabolites share structural similarities with those found in *N. antibiotica*, which also produces bioactive compounds (Stodůlková et al. 2014). Such findings suggest that members of *Nigrograna* have significant potential for biotechnological applications, particularly in drug discovery. Understanding the relationships between *Nigrograna* species and their medicinal plant hosts, as well as the impact of fungal colonization on the quality of these plants, remains a critical area of research.

In conclusion, this study highlights the diversity of *Nigrograna* species associated with medicinal plants in Southwest China and underscores the importance of integrating morphological and molecular data for accurate species identification. Given the potential ecological and economic implications of *Nigrograna* colonization on medicinal plants, continued research is essential. Detailed taxonomic and ecological studies of *Nigrograna* from medicinal plants will provide valuable insights into the species diversity, host specificity, and potential biotechnological applications of this genus. Ongoing efforts to collect and analyze fresh isolates will further enhance our understanding of the genus and its broader ecological and medicinal significance.

Acknowledgments

Hong-Zhi Du is grateful to Dr. Shaun Pennycook for corrections on the Latin names of the novel taxa. Ning-Guo Liu and Chuan-Gen Lin are thanked for their valuable suggestions on taxonomic identification. Hong-Zhi Du thanks Yong-Xiu Yu, Xiang-Dong Liang and Yue Xiao for their help with sample collections.

Additional information

Conflict of interest

The authors have declared that no competing interests exist.

Ethical statement

No ethical statement was reported.

Funding

This study was supported by the Science and Technology Fundamental Resources Investigation Program (Grant No. 2021FY100906).

Author contributions

Conceptualization: HZD, JKL. Data curation: HZD, YHL, RC. Formal analysis: HZD, YHL, JKL. Funding acquisition: JKL. Investigation: HZD, YHL. Methodology: HZD. Project administration: HZD, JKL, RC. Supervision: JKL, RC. Writing – original draft: HZD. Writing – review & editing: HZD, JKL. All authors have read and agreed to the published version of the manuscript.

Author ORCIDs

Hong-Zhi Du  <https://orcid.org/0000-0003-0350-4530>

Yu-Hang Lu  <https://orcid.org/0009-0005-9819-3182>

Ratchadawan Cheewangkoon  <https://orcid.org/0000-0001-8576-3696>

Jian-Kui Liu  <https://orcid.org/0000-0002-9232-228X>

Data availability

All of the data that support the findings of this study are available in the main text.

References

- Abtahi F, Nourani SL (2017) The most important fungal diseases associated with some useful medicinal plants. In: Ghorbanpour M, Varma A (Eds) *Medicinal Plants and Environmental Challenges*. Springer, Cham, 279–293. https://doi.org/10.1007/978-3-319-68717-9_16
- Ahmed SA, van de Sande WW, Stevens DA, Fahal A, van Diepeningen AD, Menken SB, de Hoog GS (2014) Revision of agents of black-grain eumycetoma in the order Pleosporales. *Persoonia* 33(1): 141–154. <https://doi.org/10.3767/003158514X684744>
- Ali SI, Sheikh WM, Rather MA, Venkatesalu V, Muzamil Bashir S, Nabi SU (2021) Medicinal plants: Treasure for antiviral drug discovery. *Phytotherapy Research* 35(7): 3447–3483. <https://doi.org/10.1002/ptr.7039>
- Atanasov AG, Zotchev SB, Dirsch VM, Orhan IE, Banach M, Rollinger JM, Barreca D, Weckwerth W, Bauer R, Bayer EA, Majeed M, Bishayee A, Bochkov V, Bonn GK, Braidy N, Bucar F, Cifuentes A, D’Onofrio G, Bodkin M, Diederich M, Dinkova-Kostova AT, Efferth T, El Bairi K, Arkells N, Fan TP, Fiebich BL, Freissmuth M, Georgiev MI, Gibbons S, Godfrey KM, Gruber CW, Heer J, Huber LA, Ibanez E, Kijjoo A, Kiss AK, Lu A, Macias FA, Miller MJS, Mocan A, Müller R, Nicoletti F, Perry G, Pittalà V, Rastrelli L, Ristow M, Russo GL, Silva AS, Schuster D, Sheridan H, Skalicka-Woźniak K, Skaltsounis L, Sobarzo-Sánchez E, Brecht DS, Stuppner H, Sureda A, Tzvetkov NT, Vacca RA, Aggarwal BB, Battino M, Giampieri F, Wink M, Wolfender JL, Xiao J, Yeung AWK, Lizard G, Popp MA, Heinrich M, Berindan-Neagoe I, Stadler M, Daglia M, Verpoorte R, Supuran CT (2021) Natural products in drug discovery: Advances and opportunities. *Nature Reviews. Drug Discovery* 20(3): 200–216. <https://doi.org/10.1038/s41573-020-00114-z>
- Balekundri A, Mannur V (2020) Quality control of the traditional herbs and herbal products: A review. *Future Journal of Pharmaceutical Sciences* 6(1): 67. <https://doi.org/10.1186/s43094-020-00091-5>
- Boonmee S, Wanasinghe DN, Calabon MS, Huanraluek N, Chandrasiri SKU, Jones GEB, Rossi W, Leonardi M, Singh SK, Rana S, Singh PN, Maurya DK, Lagashetti AC, Choudhary D, Dai YC, Zhao CL, Mu YH, Yuan HS, He SH, Phookamsak R, Jiang HB, Martín MP, Dueñas M, Telleria MT, Kałucka IL, Jagodziński AM, Liimatainen K, Pereira DS, Phillips AJL, Suwannarach N, Kumla J, Khuna S, Lumyong S, Potter TB, Shivas RG, Sparks AH, Vaghefi N, Abdel-Wahab MA, Abdel-Aziz FA, Li GJ, Lin WF, Singh U, Bhatt RP, Lee HB, Nguyen TTT, Kirk PM, Dutta AK, Acharya K, Sarma VV, Niranjana M, Rajeshkumar KC, Ashtekar N, Lad S, Wijayawardene NN, Bhat DJ, Xu RJ, Wijesinghe SN, Shen HW, Luo ZL, Zhang JY, Sysouphanthong P, Thongklang N, Bao DF, Aluthmuhandiram JVS, Abdollahzadeh J, Javadi A, Dovana F, Usman M, Khalid AN, Dissanayake AJ, Telagathoti A, Probst M, Peintner U, Garrido-Benavent I, Bóna L, Merényi Z, Boros L, Zoltán B, Stielow JB, Jiang N, Tian CM, Shams E, Dehghanizadeh F, Pordel A, Javan-Nikkhah M, Denchev TT, Denchev CM, Kemler M, Begerow D, Deng CY, Harrower E, Bozorov T, Kholmuradova T, Gafforov Y, Abdurazakov A, Xu JC, Mortimer PE, Ren GC, Jeewon R, Maharachchikumbura SSN, Phukhamsakda C, Mapook A, Hyde KD (2021) Fungal diversity notes 1387–1511: Taxonomic and phylogenetic contributions on genera and species of fungal taxa. *Fungal Diversity* 111(1): 1–335. <https://doi.org/10.1007/s13225-021-00489-3>
- Bundhun D, Jayawardena RS, Camporesi E (2023) Introducing *Nigrograna italica* sp. nov. (Nigrogranaceae) from *Corylus avellana* and *Valsaria tectonae* sp. nov. (Valsariaceae) from *Tectona grandis*. *Phytotaxa* 618(2): 103–119. <https://doi.org/10.11646/phytotaxa.618.2.1>

- Capella-Gutierrez S, Silla-Martinez JM, Gabaldon T (2009) TrimAl: A tool for automated alignment trimming in large-scale phylogenetic analyses. *Bioinformatics* (Oxford, England) 25(15): 1972–1973. <https://doi.org/10.1093/bioinformatics/btp348>
- Chethana KWT, Rathnayaka AR, Samarakoon BC, Wu N, Wijesinghe SN, Yasanthika WAE, Sysouphanthong P, Thiyagaraja V, Armand A, Lestari AS, Madagammana AD, Ediriweera AN, Prematunga C, Li CJ-Y, Tennakoon DS, Gomdola D, Marasinghe DS, Bundhun D, Pem D, Ren G, Zhao H, Su HL, Win H, Li H, Lu L, Calabon MS, Samarakoon MC, Chaiwan N, Huanraluek N, Samaradiwakara NP, Kularathnage ND, Abeywickrama PD, Perera RH, Tang SM, Du T-Y, Punyaboon W, Ma X, Yang YH, Tun ZL, Bhunjun CS, Manawasinghe IS, Senanayake IC, Liu JK, Tibpromma S, Wadduwage KS, Wijayalath WHDN, Raspé O, Bulgakov TS, Camporesi E, Promputtha I, Hyde KD (2023) AJOM new records and collections of fungi: 151–200. *Asian Journal of Mycology* 6(2): 89–243. <https://doi.org/10.5943/ajom/6/2/7>
- Cole I, Saxena P, Murch S (2007) Medicinal biotechnology in the genus *Scutellaria*. In *Vitro Cellular & Developmental Biology*. Plant 43(4): 318–327. <https://doi.org/10.1007/s11627-007-9055-4>
- Dai DQ, Phookamsak R, Wijayawardene NN, Li WJ, Bhat DJ, Xu JC, Taylor JE, Hyde KD, Chukeatirote E (2017) Bambusicolous fungi. *Fungal Diversity* 82(1): 1–105. <https://doi.org/10.1007/s13225-016-0367-8>
- Dayarathne MC, Jones EBG, Maharachchikumbura SSN, Devadatha B, Sarma V, Khongphinitbunjong K, Chomnunti P, Hyde KD (2020) Morpho-molecular characterization of microfungi associated with marine based habitats. *Mycosphere* 11(1): 1–188. <https://doi.org/10.5943/mycosphere/11/1/1>
- de Gruyter J, Woudenberg JH, Aveskamp MM, Verkley GJ, Groenewald JZ, Crous PW (2012) Redisposition of phoma-like anamorphs in Pleosporales. *Studies in Mycology* 75: 1–36. <https://doi.org/10.3114/sim0004>
- de Silva NI, Hyde KD, Lumyong S, Phillips AJL, Bhat DJ, Maharachchikumbura SSN, Thambugala KM, Tennakoon DS, Suwannarach N, Karunarathna SC (2022) Morphology, phylogeny, host association and geography of fungi associated with plants of Annonaceae, Apocynaceae and Magnoliaceae. *Mycosphere* 13(1): 955–1076. <https://doi.org/10.5943/mycosphere/13/1/12>
- Dissanayake AJ, Bhunjun CS, Maharachchikumbura SSN, Liu JK (2020) Applied aspects of methods to infer phylogenetic relationships amongst fungi. *Mycosphere* 11(1): 2652–2676. <https://doi.org/10.5943/mycosphere/11/1/18>
- Dong W, Wang B, Hyde KD, McKenzie EHC, Raja HA, Tanaka K, Abdel-Wahab MA, Abdel-Aziz FA, Doilom M, Phookamsak R, Hongsan S, Wanasinghe DN, Yu XD, Wang GN, Yang H, Yang J, Thambugala KM, Tian Q, Luo ZL, Yang JB, Miller AN, Fournier J, Boonmee S, Hu DM, Nalumpang S, Zhang H (2020) Freshwater Dothideomycetes. *Fungal Diversity* 105(1): 319–575. <https://doi.org/10.1007/s13225-020-00463-5>
- Du TY, Karunarathna SC, Zhang X, Dai DQ, Gao Y, Mapook A, Tibpromma S (2022a) Morphology and multigene phylogeny revealed *Peroneutypa aquilariae* sp. nov. (Diatrypaceae, Xylariales) from *Aquilaria sinensis* in Yunnan Province, China. *Studies in Fungi* 7(1): 1–11. <https://doi.org/10.48130/SIF-2022-0018>
- Du TY, Karunarathna SC, Zhang X, Dai DQ, Mapook A, Suwannarach N, Xu JC, Stephenson SL, Elgorban AM, Al-Rejaie S, Tibpromma S (2022b) Endophytic fungi associated with *Aquilaria sinensis* (Agarwood) from China show antagonism against bacterial and fungal pathogens. *Journal of Fungi* (Basel, Switzerland) 8(11): 1197. <https://doi.org/10.3390/jof8111197>
- Guo B, Luo W, Zang W (2020) Spatial-temporal shifts of ecological vulnerability of Karst Mountain ecosystem-impacts of global change and anthropogenic interference.

- The Science of the Total Environment 741: 140256. <https://doi.org/10.1016/j.scitotenv.2020.140256>
- Helaly SE, Thongbai B, Stadler M (2018) Diversity of biologically active secondary metabolites from endophytic and saprotrophic fungi of the ascomycete order Xylariales. Natural Product Reports 35(9): 992–1014. <https://doi.org/10.1039/C8NP00010G>
- Hongsanan S, Hyde KD, Phookamsak R, Wanasinghe DN, McKenzie EHC, Sarma VV, Boonmee S, Lücking R, Bhat DJ, Liu NG, Tennakoon DS, Pem D, Karunarathna A, Jiang SH, Jones EBG, Phillips AJL, Manawasinghe IS, Tibpromma S, Jayasiri SC, Sandamali DS, Jayawardena RS, Wijayawardene NN, Ekanayaka AH, Jeewon R, Lu YZ, Dissanayake AJ, Zeng XY, Luo ZL, Tian Q, Phukhamsakda C, Thambugala KM, Dai DQ, Chethana KWT, Samarakoon MC, Ertz D, Bao DF, Doilom M, Liu JK, PérezOrtega S, Suija A, Senwanna C, Wijesinghe SN, Konta S, Niranjana M, Zhang SN, Ariyawansa HA, Jiang HB, Zhang JF, Norphanphoun C, de Silva NI, Thiagaraja V, Zhang H, Bezerra JDP, Miranda-González R, Aptroot A, Kashiwadani H, Harishchandra D, Sérusiaux E, Aluthmuhandiram JVS, Abeywickrama PD, Devadatha B, Wu HX, Moon KH, Gueidan C, Schumm F, Bundhun D, Mapook A, Monkai J, Chomnunti P, Suetrong S, Chaiwan N, Dayarathne MC, Yang J, Rathnayaka AR, Bhunjun CS, Xu JC, Zheng JS, Liu G, Feng Y, Xie N (2020) Refined families of Dothideomycetes: Dothideomycetidae and Pleosporomycetidae. Mycosphere 11(1): 1553–2107. <https://doi.org/10.5943/mycosphere/11/1/13>
- Hu HM, He MH, Wu YP, Long SH, Zhang X, Liu LL, Shen XC, Wijayawardene NN, Meng ZB, Long QD, Kang JC, Li QR (2023) Taxonomic and phylogenetic characterisations of six species of Pleosporales (in Didymosphaeriaceae, Roussellaceae and Nigrogranaeae) from China. MycoKeys 100: 123–151. <https://doi.org/10.3897/mycokeys.100.109423>
- Huang LQ, Xiao PG, Wang YY (2012) Investigation on resources of rare and endangered medicinal plants in China. Shanghai Scientific and Technical Publishers, Shanghai, 284 pp.
- Hyde KD, Norphanphoun C, Abreu VP, Bazzicalupo A, Thilini Chethana KW, Clericuzio M, Dayarathne MC, Dissanayake AJ, Ekanayaka AH, He MQ, Hongsanan S, Huang SK, Jayasiri SC, Jayawardena RS, Karunarathna A, Konta S, Kušan I, Lee H, Li J, Lin CG, Liu NG, Lu YZ, Luo ZL, Manawasinghe IS, Mapook A, Perera RH, Phookamsak R, Phukhamsakda C, Siedlecki I, Soares AM, Tennakoon DS, Tian Q, Tibpromma S, Wanasinghe DN, Xiao YP, Yang J, Zeng XY, Abdel-Aziz FA, Li WJ, Senanayake IC, Shang QJ, Daranagama DA, de Silva NI, Thambugala KM, Abdel-Wahab MA, Bahkali AH, Berbee ML, Boonmee S, Bhat DJ, Bulgakov TS, Buyck B, Camporesi E, Castañeda-Ruiz RF, Chomnunti P, Doilom M, Dovana F, Gibertoni TB, Jadan M, Jeewon R, Jones EBG, Kang JC, Karunarathna SC, Lim YW, Liu JK, Liu ZY, Plautz Jr HL, Lumyong S, Maharachchikumbura SSN, Matočec N, McKenzie EHC, Mešić A, Miller D, Pawłowska J, Pereira OL, Promputtha I, Romero AI, Ryvarden L, Su HY, Suetrong S, Tkáčec Z, Vizzini A, Wen TC, Wisitrassameewong K, Wrzosek M, Xu JC, Zhao Q, Zhao RL, Mortimer PE (2017) Fungal diversity notes 603–708: Taxonomic and phylogenetic notes on genera and species. Fungal Diversity 87(1): 1–235. <https://doi.org/10.1007/s13225-017-0391-3>
- Hyde KD, Norphanphoun C, Ma J, Yang HD, Zhang JY, Du TY, Gao Y, Gomes de Farias AR, Gui H, He SC, He YK, Li CJY, Liu XF, Lu L, Su HL, Tang X, Tian XG, Wang SY, Wei DP, Xu RF, Xu RJ, Yang Q, Yang YY, Zhang F, Zhang Q, Bahkali AH, Boonmee S, Chethana KWT, Jayawardena RS, Lu YZ, Karunarathna SC, Tibpromma S, Wang Y, Zhao Q (2023) Mycosphere notes 387–412 – novel species of fungal taxa from around the world. Mycosphere 14(1): 663–744. <https://doi.org/10.5943/mycosphere/14/1/8>
- Jaklitsch WM, Voglmayr H (2016) Hidden diversity in *Thyridaria* and a new circumscription of the Thyridariaceae. Studies in Mycology 85(1): 35–64. <https://doi.org/10.1016/j.simyco.2016.09.002>

- Katoh K, Standley DM (2013) MAFFT multiple sequence alignment software version 7: Improvements in performance and usability. *Molecular Biology and Evolution* 30(4): 772–780. <https://doi.org/10.1093/molbev/mst010>
- Keshri P, Rai N, Verma A, Kamble S, Barik S, Mishra P, Singh S, Salvi P, Gautam V (2021) Biological potential of bioactive metabolites derived from fungal endophytes associated with medicinal plants. *Mycological Progress* 20(5): 577–594. <https://doi.org/10.1007/s11557-021-01695-8>
- Kolařík M (2018) New taxonomic combinations in endophytic representatives of the genus *Nigrograna*. *Czech Mycology* 70(2): 123–126. <https://doi.org/10.33585/cmy.70202>
- Kolařík M, Spakowicz DJ, Gazis R, Shaw J, Kubátová A, Nováková A, Chudíčková M, Forcina GC, Kang KW, Kelnarová I, Skaltsas D, Portero CE, Strobel SA, Narváez-Trujillo A (2017) *Biatriospora* (Ascomycota: Pleosporales) is an ecologically diverse genus including facultative marine fungi and endophytes with biotechnological potential. *Plant Systematics and Evolution* 303(1): 35–50. <https://doi.org/10.1007/s00606-016-1350-2>
- Larget B, Simon DL (1999) Markov chain Monte Carlo algorithms for the Bayesian analysis of phylogenetic trees. *Molecular Biology and Evolution* 16(6): 750–759. <https://doi.org/10.1093/oxfordjournals.molbev.a026160>
- Larsson A (2014) AliView: A fast and lightweight alignment viewer and editor for large datasets. *Bioinformatics (Oxford, England)* 30(22): 3276–3278. <https://doi.org/10.1093/bioinformatics/btu531>
- Li WL, Liang RR, Dissanayake AJ, Liu JK (2023) Mycosphere Notes 413–448: Dothideomycetes associated with woody oil plants in China. *Mycosphere* 14(1): 1436–1529. <https://doi.org/10.5943/mycosphere/14/1/16>
- Liu YJ, Whelen S, Hall BD (1999) Phylogenetic relationships among Ascomycetes: Evidence from an RNA polymerase II subunit. *Molecular Biology and Evolution* 16(12): 1799–1808. <https://doi.org/10.1093/oxfordjournals.molbev.a026092>
- Liu JK, Chomnunti P, Cai L, Phookamsak R, Chukeatirote R, Jones EBG, Moslem M, Hyde KD (2010) Phylogeny and morphology of *Neodeightonia palmicola* sp. nov. from palms. *Sydowia* 62(2): 261–276. https://www.zobodat.at/pdf/Sydowia_062_0261-0276.pdf
- Liu JK, Hyde KD, Jeewon R, Phillips AJ, Maharachchikumbura SS, Ryberg M, Liu ZY, Zhao Q (2017) Ranking higher taxa using divergence times: A case study in Dothideomycetes. *Fungal Diversity* 84(1): 75–99. <https://doi.org/10.1007/s13225-017-0385-1>
- Liu SL, Wang XW, Li GJ, Deng CY, Rossi W, Leonardi M, Liimatainen K, Kekki T, Niskanen T, Smith ME, Ammirati J, Bojantchev D, Abdel-Wahab MA, Zhang M, Tian E, Lu YZ, Zhang JY, Ma J, Dutta AK, Acharya K, Du TY, Xu J, Kim JS, Lim YW, Gerlach A, Zeng NK, Han YX, Razaghi P, Raza M, Cai L, Calabon MS, Jones EBG, Saha R, Kumar TKA, Krishnapriya K, Thomas A, Kaliyaperumal M, Kezo K, Gunaseelan S, Singh SK, Singh PN, Lagashetti AC, Pawar KS, Jiang S, Zhang C, Zhang H, Qing Y, Bau T, Peng XC, Wen TC, Ramirez NA, Niveiro N, Li MX, Yang ZL, Wu G, Tarafder E, Tennakoon DS, Kuo CH, da Silva TM, Souza-Motta CM, Bezerra JDP, He G, Ji XH, Suwannarach N, Kumla J, Lumyong S, Wannathes N, Rana S, Hyde KD, Zhou LW (2024) Fungal diversity notes 1717–1817: Taxonomic and phylogenetic contributions on genera and species of fungal taxa. *Fungal Diversity* 124(1): 1–216. <https://doi.org/10.1007/s13225-023-00529-0>
- Lu LM, Mao LF, Yang T, Ye JF, Liu B, Li HL, Sun M, Miller JT, Mathews S, Hu HH, Niu YT, Peng DX, Chen YH, Smith SA, Chen M, Xiang KL, Le CT, Dang VC, Lu AM, Soltis PS, Soltis DE, Li JH, Chen ZD (2018) Evolutionary history of the angiosperm flora of China. *Nature* 554(7691): 234–238. <https://doi.org/10.1038/nature25485>
- Lu L, Karunarathna SC, Dai DQ, Jayawardena RS, Suwannarach N, Tibpromma S (2022) Three new species of *Nigrograna* (Dothideomycetes, Pleosporales) associated with

- Arabica coffee* from Yunnan Province, China. MycoKeys 94: 51–71. <https://doi.org/10.3897/mycokeys.94.95751>
- Mack JN, Sproule A, Shields SW, Seifert KA, Smith M, Overy DP (2024) Two novel Pleosporales species isolated from the bark of *Acer saccharum*. Fungal Systematics and Evolution 13(1): 1–14. <https://doi.org/10.3114/fuse.2024.13.01>
- Mapook A, Hyde KD, Hyde KD, Hyde KD, McKenzie EHC, Jones EBG, Bhat DJ, Jeewon R, Stadler M, Samarakoon MC, Malaithong M, Tanunchai B, Buscot F, Wubet T, Purahong WJFD (2020) Taxonomic and phylogenetic contributions to fungi associated with the invasive weed *Chromolaena odorata* (Siam weed). Fungal Diversity 101(1): 1–175. <https://doi.org/10.1007/s13225-020-00444-8>
- Miller MA, Pfeiffer W, Schwartz T (2010) Creating the CIPRES science gateway for inference of large phylogenetic trees. Proceedings of the Gateway Computing Environments Workshop (GCE) 14: 1–8. <https://doi.org/10.1109/GCE.2010.5676129>
- Nalawade S, Sagare A, Lee CY, Kao CL, Tsay HS (2003) Studies on tissue culture of Chinese medicinal plant resources in Taiwan and their sustainable utilization. Botanical Bulletin-Academia Sinica Taipei 44(2): 79–98. <https://ejournal.sinica.edu.tw/bbas/content/2003/2/bot442-01.pdf>
- Naysmith BJ, Hume PA, Sperry J, Brimble MA (2017) Pyranonaphthoquinones - isolation, biology and synthesis: An update. Natural Product Reports 34(1): 25–61. <https://doi.org/10.1039/C6NP00080K>
- Nylander J (2008) MrModeltest 2 v. 2.3 (Program for selecting DNA substitution models using PAUP*). Evolutionary Biology Centre, Uppsala, Sweden. <https://mybiosoftware.com/mrmodeltest-2-3-program-selecting-dna-substitution-models-paup.html>
- Rahman IU, Afzal A, Iqbal Z, Ijaz F, Ali N, Shah M, Ullah S, Bussmann RW (2019) Historical perspectives of ethnobotany. Clinics in Dermatology 37(4): 382–388. <https://doi.org/10.1016/j.clindermatol.2018.03.018>
- Rannala B, Yang Z (1996) Probability distribution of molecular evolutionary trees: A new method of phylogenetic inference. Journal of Molecular Evolution 43(3): 304–311. <https://doi.org/10.1007/BF02338839>
- Rasool A, Bhat KM, Sheikh AA, Jan A, Hassan S (2020) Medicinal plants: Role, distribution and future. Journal of Pharmacognosy and Phytochemistry 9: 2111–2114. <https://doi.org/10.15171/jhp.2018.01>
- Rehner SA, Buckley E (2005) A *Beauveria* phylogeny inferred from nuclear ITS and EF1- α sequences: Evidence for cryptic diversification and links to *Cordyceps* teleomorphs. Mycologia 97(1): 84–98. <https://doi.org/10.3852/mycologia.97.1.84>
- Ren GC, Jayasiri SC, Tibpromma S, De Farias ARG, Chethana KWT, Faraj KH, Wanasinghe DN, Xu JC, Hyde KD, Gui H (2024) Saprobic ascomycetes associated with woody litter from the Greater Mekong Subregion (Southwestern China and Northern Thailand). Mycosphere 15(1): 954–1082.
- Ronquist F, Teslenko M, van der Mark P, Ayres DL, Darling A, Hohna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP (2012) MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology 61(3): 539–542. <https://doi.org/10.1093/sysbio/sys029>
- Samy RP, Gopalakrishnakone P (2007) Current status of herbal and their future perspectives. Nature Precedings, 1–13. <https://doi.org/10.1038/npre.2007.1176.1>
- Schmidt BM (2017) Ethnobotany: a phytochemical perspective. Ethnobotany, 1–109. <https://doi.org/10.1002/9781118961933.ch1>
- Senanayake IC, Rathnayaka AR, Sandamali DS, Calabon MS, Gentekaki E, Lee HB, Hurdeal VG, Pem D, Dissanayake LS, Wijesinghe SN, Bundhun D, Nguyen TT, Goonasekara ID,

- Abeywickrama PD, Bhunjun CS, Jayawardena RS, Wanasinghe DN, Jeewon R, Bhat DJ, Xiang MM (2020) Morphological approaches in studying fungi: Collection, examination, isolation, sporulation and preservation. *Mycosphere* 11(1): 2678–2754. <https://doi.org/10.5943/mycosphere/11/1/20>
- Senanayake IC, Rossi W, Leonardi M, Weir A, McHugh M, Rajeshkumar KC, Verma RK, Karunarathna SC, Tibpromma S, Ashtekar N, Ashtamoorthy SK, Raveendran S, Kour G, Singh A, De la Peña-Lastra S, Mateos A, Kolařík M, Antonín V, Ševčíková H, Esteve-Raventós F, Larsson E, Pancorbo F, Moreno G, Altés A, Turégano Y, Du TY, Lu L, Li QR, Kang JC, Gunaseelan S, Kezo K, Kaliyaperumal M, Fu J, Samarakoon MC, Gafforov Y, Teshaboeva S, Kunjan PC, Chamaparambath A, Flakus A, Etayo J, Rodriguez-Flakus P, Zhurbenko MP, de Silva NI, Tennakoon DS, Latha KPD, Manimohan P, Raj KNA, Calabon MS, Ahmadpour A, Heidarian Z, Alavi Z, Alavi F, Ghosta Y, Azizi R, Luo M, Zhao MP, Kularathnage ND, Hua L, Yang YH, Liao CF, Zhao HJ, Lestari AS, Jayasiri SC, Yu FM, Lei L, Liu JW, Karimi O, Tang SM, Sun YR, Wang Y, Zeng M, Htet ZH, Linaldeddu BT, Alves A, Phillips AJL, Bregant C, Montecchio L, De Kesel A, Hustad VP, Miller AN, Fedosova AG, Kučera V, Raza M, Hussain M, Chen YP, Thiyagaraja V, Gomdola D, Rathnayaka AR, Dissanayake AJ, Suwannarach N, Hongsan S, Maharachchikumbura SSN, Dissanayake LS, Wijayawardene NN, Phookamsak R, Lumyong S, Jones EBG, Yapa N, Wanasinghe DN, Xie N, Doilom M, Manawasinghe IS, Liu JK, Zhao Q, Xu B, Hyde KD, Song J (2023) Fungal diversity notes 1611–1716: Taxonomic and phylogenetic contributions on fungal genera and species emphasis in south China. *Fungal Diversity* 122(1): 161–403. <https://doi.org/10.1007/s13225-023-00523-6>
- Shan ZJ, Ye JF, Hao DC, Xiao PG, Chen ZD, Lu AM (2022) Distribution patterns and industry planning of commonly used traditional Chinese medicinal plants in China. *Plant Diversity* 44(3): 255–261. <https://doi.org/10.1016/j.pld.2021.11.003>
- Shu YX, Cheng R, Huang J, Li K, Zhao S, Zhao H, Dong W, Dong Z, Doilom M, Xu B (2023) Introducing *Nigrograna wuhanensis* sp. nov. (Nigrogranaceae, Pleosporales) from *Camellia japonica* in China. *Warasan Khana Witthayasat Maha Witthayalai Chiang Mai* 50(3): 1–11. <https://doi.org/10.12982/CMJS.2023.025>
- Stamatakis A (2006) RAxML-VI-HPC: Maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics (Oxford, England)* 22(21): 2688–2690. <https://doi.org/10.1093/bioinformatics/btl446>
- Stamatakis A, Hoover P, Rougemont J (2008) A rapid bootstrap algorithm for the RAxML web servers. *Systematic Biology* 57(5): 758–771. <https://doi.org/10.1080/10635150802429642>
- Stodůlková E, Man P, Kuzma M, Černý J, Císařová I, Kubátová A, Chudíčková M, Kolařík M, Flieger M (2014) A highly diverse spectrum of naphthoquinone derivatives produced by the endophytic fungus *Biatrispora* sp. CCF 4378. *Folia Microbiologica* 60(3): 259–267. <https://doi.org/10.1007/s12223-014-0366-7>
- Sun YR, Jayawardena RS, Sun JE, Wang Y (2023) Pestalotioid species associated with medicinal plants in Southwest China and Thailand. *Microbiology Spectrum* 11(1): e0398722. <https://doi.org/10.1128/spectrum.03987-22>
- Taylor DL, Hollingsworth T, McFarland J, Lennon N, Nusbaum C, Ruess R (2014) A first comprehensive census of fungi in soil reveals both hyperdiversity and fine-scale niche partitioning. *Ecological Monographs* 84(1): 3–20. <https://doi.org/10.1890/12-1693.1>
- Tibpromma S, Hyde KD, Jeewon R, Maharachchikumbura SSN, Liu JK, Bhat DJ, Jones EBG, McKenzie EHC, Camporesi E, Bulgakov TS, Doilom M, de Azevedo Santiago

- ALCM, Das K, Manimohan P, Gibertoni TB, Lim YW, Ekanayaka AH, Thongbai B, Lee HB, Yang JB, Kirk PM, Sysouphanthong P, Singh SK, Boonmee S, Dong W, Raj KNA, Latha KPD, Phookamsak R, Phukhamsakda C, Konta S, Jayasiri SC, Norphanphoun C, Tennakoon DS, Li J, Dayarathne MC, Perera RH, Xiao Y, Wanasinghe DN, Senanayake IC, Goonasekara ID, de Silva NI, Mapook A, Jayawardena RS, Dissanayake AJ, Manawasinghe IS, Chethana KWT, Luo ZL, Hapuarachchi KK, Baghela A, Soares AM, Vizzini A, Meiras-Ottoni A, Mešić A, Dutta AK, de Souza CAF, Richter C, Lin CG, Chakrabarty D, Daranagama DA, Lima DX, Chakraborty D, Ercole E, Wu F, Simonini G, Vasquez G, da Silva GA, Plautz HL, Ariyawansa HA, Lee H, Kušan I, Song J, Sun J, Karmakar J, Hu K, Semwal KC, Thambugala KM, Voigt K, Acharya K, Rajeshkumar KC, Ryvarden L, Jadan M, Hosen MI, Mikšić M, Samarakoon MC, Wijayawardene NN, Kim NK, Matočec N, Singh PN, Tian Q, Bhatt RP, de Oliveira RJV, Tulloss RE, Aamir S, Kaewchai S, Marathe SD, Khan S, Hongsanant S, Adhikari S, Mehmood T, Bandyopadhyay TK, Svetasheva TY, Nguyen TTT, Antonín V, Li WJ, Wang Y, Indoliya Y, Tkalčec Z, Elgorban AM, Bahkali AH, Tang AMC, Su HY, Zhang H, Promptutha I, Luangsa-ard J, Xu JC, Yang J, Kang JC, Stadler M, Mortimer PE, Chomnunti P, Zhao Q, Phillips AJL, Nontachaiyapoom S, Wen TC, Karunarathna SC (2017) Fungal diversity notes 491–602: Taxonomic and phylogenetic contributions to fungal taxa. *Fungal Diversity* 83(1): 1–261. <https://doi.org/10.1007/s13225-017-0378-0>
- Vaidya G, Lohman DJ, Meier R (2011) SequenceMatrix concatenation software for the fast assembly of multi gene datasets with character set and codon information. *Cladistics* 27(2): 171–180. <https://doi.org/10.1111/j.1096-0031.2010.00329.x>
- Vilgalys R, Hester M (1990) Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *Journal of Bacteriology* 172(8): 4238–4246. <https://doi.org/10.1128/jb.172.8.4238-4246.1990>
- Wanasinghe DN, Wijayawardene NN, Xu JC, Cheewangkoon R, Mortimer PE (2020) Taxonomic novelties in *Magnolia*-associated pleosporalean fungi in the Kunming Botanical Gardens (Yunnan, China). *PLoS ONE* 15(7): e0235855. <https://doi.org/10.1371/journal.pone.0235855>
- White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. *PCR protocols: a guide to methods and applications* 18(1): 315–322. <https://doi.org/10.1016/B978-0-12-372180-8.50042-1>
- Wu N, Thilini Chethana KW, Liu JK (2024) *Velutinus*, a novel genus in Patellariopsidaceae from medicinal plants in Sichuan Province, China. *Phytotaxa* 638(2): 165–174. <https://doi.org/10.11646/phytotaxa.638.2.5>
- Xu RF, Karunarathna SC, Phukhamsakda C, Dai DQ, Elgorban AM, Suwannarach N, Kumla J, Wang XY, Tibpromma S (2024) Four new species of Dothideomycetes (Ascomycota) from Pará Rubber (*Hevea brasiliensis*) in Yunnan Province, China. *Mycosphaera* 103: 71–95. <https://doi.org/10.3897/mycokeys.103.117580>
- Zhang JF, Liu JK, Thambugala KM, Yang J, Meng ZH, Liu ZY (2020) Two new species and a new record of *Nigrograna* (Nigrogranaceae, Pleosporales) from China and Thailand. *Mycological Progress* 19(11): 1365–1375. <https://doi.org/10.1007/s11557-020-01633-0>
- Zhang ZY, Pan H, Tao G, Li X, Han YF, Feng Y, Tong SQ, Ding CY (2024) Culturable Mycobiota from Guizhou Wildlife Park in China. *Mycosphere* 15(1): 654–763.
- Zhao YZ, Zhang ZF, Cai L, Peng WJ, Liu F (2018) Four new filamentous fungal species from newly-collected and hive-stored bee pollen. *Mycosphere* 9(6): 1089–1116. <https://doi.org/10.5943/mycosphere/9/6/3>